#18

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re: United States Patent No. 5,951,974	
	Attn: Box Patent Ext.
Inventor: Carl W. Gilbert, et al.	
Issue Date: September 14, 1999	

Honorable Commissioner of Patents
and Trademarks
Washington, D.C. 20231

LETTER OF TRANSMITTAL OF APPLICATION FOR EXTENSION OF PATENT TERM

Sir:

Transmitted herewith for filing is an application for extension of term of U.S. Patent No. 5,951,974 and a duplicate of the papers thereof, certified as such.

Also submitted herewith is an additional original declaration for extension of U.S. Patent No. 5,951,974. Therefore, the present application is complete.

Applicant, Schering Corporation ("Schering") states that Schering is the authorized agent for Enzon, Inc. ("Enzon") owner of U.S. Patent No. 5,951,974 (see Exhibit I); that Schering is the holder of the regulatory approval granted with respect to the regulatory review period for PEG-Intron™ (Peginterferon alfa-2b) Powder for Injection as evidenced by: (1) submission on June 2, 1997 by Schering of BB-IND No. 7173 to the Food

and Drug Administration ("FDA") for the purpose of conducting clinical studies for the use of Pegylated Interferon Alfa-2b (SCH 54031) Powder for Injection for use in humans (see Exhibit IV); (2) the submission on December 22, 1999 by Schering of BLA No. 19-1488 (see Exhibit VI); and (3) the FDA letter dated January 19, 2001 approving BLA No. 103949 (replaces Ref. BLA No. 99-1488 for peginterferon alfa-2b for treatment of chronic hepatitis in patients not previously treated with interferon alfa who have compensated liver disease and at least 18 years of age. See Exhibits VII & XII.

The Commissioner is hereby authorized to charge payment in the amount of $ 1,120.00 and of any additional fees associated with this communication or credit any overpayment to Deposit Account No. 19-0365. A duplicate copy of this sheet is enclosed.

Respectfully submitted,

Thomas D. Hoffman
Registration No. 28221
Attorney for Authorized Agent of the
Assignee of Record
Telephone No. (908) 298-5037

SCHERING-PLOUGH CORPORATION
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Kenilworth, New Jersey 07033-0530

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

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In re: United States Patent No. 5,951,974 :

: Attn: Box Patent Ext.

Inventor: Carl W. Gilbert, et al. :

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Honorable Commissioner of Patents
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Washington, D.C. 20231

REQUEST FOR EXTENSION OF PATENT TERM UNDER 35 U.S.C. §156

Sir:

Pursuant to 35 U.S.C. §156 and 37 C.F.R. §§1.701-1.791, Schering Corporation ("Schering"), authorized agent (see Exhibit I) for Enzon, Inc. ("Enzon"), owner of the above-identified patent by virtue of the following Assignments (I) The Assignment by Myung-ok Park-Cho (executed on November 11, 1997), of his interests in Serial No. 08/994,622, filed December 19, 1997 which issued as the above-identified patent, said Assignment being recorded in the United States Patent and Trademark Office ("USPTO") on December 22, 1997 at Reel 9063, Frame 0929 and attached hereto as Exhibit VIII; (2) The Assignment by Carl W. Gilbert (executed on November 7, 1997) of his interest in Serial No. 08/994,622, filed December 19, 1997 which issued as the above identified patent, said Assignment being recorded in the USPTO on March 6, 1998 at Reel 9063, Frame 0961; and attached hereto as Exhibit IX hereby requests an extension of the 20 year from filing

date patent term set pursuant to 35 U.S.C. §154(a)(2) of United States Patent No. 5,951,974.

The following information is submitted in accordance with 35 U.S.C. §156(d) and the rules for extension of patent term issued by the USPTO at 37 C.F.R. Subpart F, §§1.701 to 1.791 and follows the numerical format set forth in 37 C.F.R. §1.740:

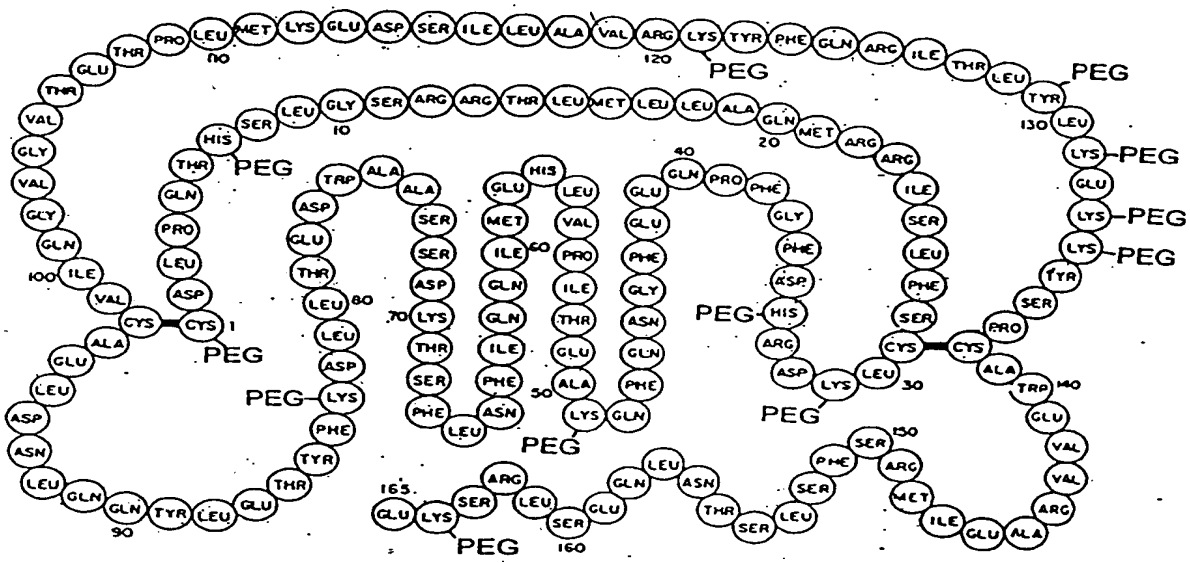
(1) A COMPLETE IDENTIFICATION OF THE APPROVED PRODUCT AS BY APPROPRIATE CHEMICAL AND GENERIC NAMES, PHYSICAL STRUCTURE OR CHARACTERISTICS:

The approved product is PEG-Intron™ (Peginterferon alfa-2b) Powder for Injection. As used herein, the USAN name for the active ingredient in the approved product is peginterferon alfa-2b. As shown in Exhibit II (Section 4.A.1., page 1 and Section 4.A.1.2. pages 4,7, 36 and 94 and 95 of Description/Characterization of BLA 99-1488), the active ingredient in the approved product has the following physical and chemical description and chemical names and chemical structural formula:

Physical Description:

Clear to opalescent, colourless to slightly yellow solution; essentially free of visible particles.

Chemical Structure:



Note: The predominant form is one PEG molecule attached to each IFN molecule; all positional isomers of the pegylated product have been identified and include attachment of one PEG molecule at any of 12 different sites including 8 Lysines, 2 Histidines, 1 Tyrosine, and 1 N-terminal Cysteine.

Molecular Weight:

Approximately 31,870 daltons (due to PEG polymer heterogeneity)

USAN Name:	peginterferon alfa-2b
Code Name:	SCH 54031
CAS Number:	215647-85-1
Other Names:	PEG-IFN
	PEG-Interferon alfa-2b
	PEG-Intron™ (commercial product)

(2) A COMPLETE IDENTIFICATION OF THE FEDERAL STATUTE INCLUDING THE APPLICABLE PROVISION OF LAW UNDER WHICH THE REGULATORY REVIEW OCCURRED:

The regulatory review occurred under Section 351 of the Public Health Service Act, 42 U.S.C. § 262. See Exhibit VII. Section 351 of the Public Health Service Act, provides for the submission and approval of an original Biologics License Applications ("BLAs") for biologics meeting the definition of "biological product" under 42 U.S.C. §262(i).

(3) AN IDENTIFICATION OF THE DATE ON WHICH THE PRODUCT RECEIVED PERMISSION FOR COMMERCIAL MARKETING OR USE UNDER THE PROVISION OF LAW UNDER WHICH THE APPLICABLE REGULATORY REVIEW PERIOD OCCURRED:

PEG-Intron™ (peginterferon alfa-2b) powder for injection was approved by the FDA for commercial marketing on January 19, 2001 for treatment of chronic hepatitis C in patients not previously treated with interferon alfa who have compensated liver disease and at least 18 years of age. See Exhibits VII & XII.

(4) IN THE CASE OF A DRUG PRODUCT, AN IDENTIFICATION OF EACH ACTIVE INGREDIENT IN THE PRODUCT AND AS TO EACH ACTIVE INGREDIENT, A STATEMENT THAT IT HAS NOT BEEN PREVIOUSLY APPROVED FOR COMMERCIAL MARKETING OR USE UNDER THE FFDCA, THE PUBLIC HEALTH SERVICE ACT OR THE

VIRUS-SERUM TOXIN ACT OR A STATEMENT OF WHEN THE ACTIVE INGREDIENT WAS APPROVED FOR COMMERCIAL MARKETING OR USE (EITHER ALONE OR IN COMBINATION WITH OTHER ACTIVE INGREDIENTS), THE USE FOR WHICH IT WAS APPROVED, AND THE PROVISION OF LAW UNDER WHICH IT WAS APPROVED.

The active ingredient in the approved product, Peg-Intron™ (peginterferon alfa-2b) Powder for Injection, has the USAN name of peginterferon alfa-2b and the other names listed in Paragraph No. (1) hereinabove as well as in Exhibits II and XIII. The active ingredient, peginterferon alfa-2b, approved for marketing under Section 351 of the Public Health Service Act [42 U.S.C. §262 (a)(2)(B)], has not previously been approved for commercial marketing or use under the Federal Food, Drug and Cosmetic Act ("FFDCA"), The Public Health Service Act or the Virus-Serum Toxin Act. See Exhibit VII (BLA approval on January 19, 2001).

(5) A STATEMENT THAT THE APPLICATION IS BEING SUBMITTED WITHIN THE SIXTY DAY PERIOD PERMITTED FOR SUBMISSION PURSUANT TO SEC. 1.720(f) AND AN IDENTIFICATION OF THE DATE OF THE LAST DAY ON WHICH THE APPLICATION COULD BE SUBMITTED:

The product was approved on January 19, 2001, and the last day within the sixty day period permitted for submission of an application for extension of the relevant U.S. Patent is March 19, 2001. This application is being timely filed before the expiration of the March 19, 2001 deadline, pursuant to 35 U.S.C. §21(a) and (b) and 37 C.F.R. §1.7 and 1.741(a).

The first maintenance fee for U.S. Patent No. 5,951,974 is not due until March 14, 2003.

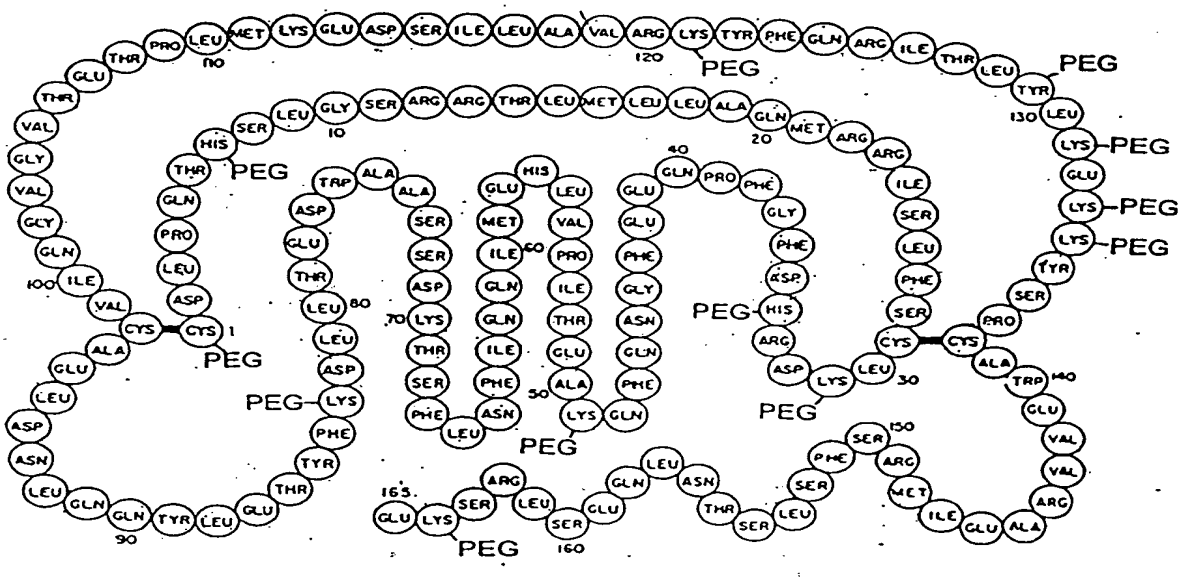
(9) A STATEMENT THAT THE PATENT CLAIMS, THE APPROVED PRODUCT OR A METHOD OF USING OR MANUFACTURING THE APPROVED PRODUCT, AND A SHOWING WHICH LISTS EACH APPLICABLE PATENT CLAIM AND DEMONSTRATES THE MANNER IN WHICH EACH APPLICABLE PATENT CLAIM READS ON THE APPROVED PRODUCT OR A METHOD OF USING OR MANUFACTURING THE APPROVED PRODUCT:

At least claims 1-11, 13-18, 34-36, 38-45 and 47 and 48 and of United States Patent No. 5,951,974 read on FDA approved product PEG-Intron™ (peginterferon alfa-2b) powder for injection for the treatment of chronic hepatitis C in patients not previously treated with interferon alfa who have compensated liver disease and are at least 18 years of age. See Product Information Sheet entitled "Description" (the paragraph in the upper left hand column of page 1) and "Indications and Usage" (second paragraph from top of left right hand column on page 2) (Exhibit XIII). See also Exhibit II, pages 1 and 2 and United States Patent No. 5,951,974, column 10, lines 28-31 and 49-54.

Claim 1 of United States Patent No. 5,951,974 reads:

1. A pharmaceutical composition, comprising a mixture of alpha interferon polymer conjugate positional isomers, wherein one of said positional isomers comprises an alpha interferon covalently conjugated to a substantially non-antigenic polymer at a histidine residue on said alpha interferon, wherein said substantially non-antigenic polymer is a polyalkylene oxide comprising an alkyl terminal.

Peginterferon alfa-2b (SCH 54031), the active ingredient in the approved product, PEG-Intron™ Powder for Injection, has the chemical structural formula set forth in paragraph 1 (page 3 hereinabove) and in Exhibit II:



Note: The predominant form is one PEG molecule attached to each IFN molecule; all positional isomers of the pegylated product have been identified and include attachment of one PEG molecule at any of 12 different sites including 8 Lysines, 2 Histidines, 1 Tyrosine, and 1 N-terminal Cysteine.

As stated in Exhibit II, Section 4.A.1 pages 1 and 4, SCH 54031 PEG_{12000} Interferon alfa-2b (IFN) is a covalent conjugate of recombinant interferon alfa-2b with monomethoxypolyethylene glycol. The predominant form is one PEG molecule attached to each IFN molecule including attachment of one PEG molecule at any of 12 different sites including 2 histidines.

Claim 1 covers peginterferon alfa-2b in that it is a mixture of alfa interferon conjugate positional isomers wherein at least one of said positional isomers comprises an alfa interferon covalently conjugated to a substantially non-antigenic polymer at a histidine residue on said interferon alfa and wherein the substantially non-antigenic polymer, a polyalkylene oxide comprising an alkyl terminal, is monomethoxypolyethylene glycol.

Claim 2 of U.S. Patent No. 5,951,974 is:

2. The pharmaceutical composition of claim 1, wherein said alpha interferon is interferon alpha 2b. See discussion regarding claim 1 and Exhibits II and XII.

Claim 2 covers peginterferon alfa-2b in that the interferon alfa is interferon alfa-2b.

Claim 3 of U.S. Patent No. 5,951,974 is:

3. The pharmaceutical composition of claim 2, wherein said histidine residue is His34.

Claim 3 covers peginterferon alfa-2b in that one of the PEG molecules is attached to one peginterferon alfa-2b at Histidine 34. See Exhibit II 4.A.1. pages 1 and 95.

Claims 4-6 of U.S. Patent No. 5,951,974 are:

4. The pharmaceutical composition of claim 1, wherein said mixture of said alpha interferon positional isomers comprises at least about 3 positional isomers.

5. The pharmaceutical composition of claim 4, wherein said mixture of said alpha interferon positional isomers comprises at least about 6 positional isomers.

6. The pharmaceutical composition of claim 5, wherein said mixture of said alpha interferon positional isomers comprises at least about 8 positional isomers.

Claims 4-6 cover peginterferon alfa-2b in that it is a mixture of alfa interferon positional isomers comprising at least 12 positional isomers. See Exhibit II at 4.A.1 page 1.

Claim 7 of U.S. Patent No. 5,951,974 is:

7. The pharmaceutical composition of claim 6, wherein said alpha interferon is alpha interferon 2b and said mixture of positional isomers comprises a substantially non-antigenic polymer linked to said alpha interferon 2b, at an amino acid residue selected from the group consisting of Cys1, Lys31, His34, Lys49, Lys83, Lys121, Lys131 and Lys134.

Claim 7 covers peginterferon alfa-2b in that the mixture of the 8 positional isomers comprises antigenic polymer linked to interferon alfa-2b at the eight (8) amino acid residues as shown in the Exhibit II (chemical structural formula) Section 4.A.1 on page 1 and the Table on page 95.

Claims 8-11 of U.S. Patent No. 5,951,974 are:

8. The pharmaceutical composition of claim 1, wherein said polyalkylene oxide is a polyethylene glycol.

9. The pharmaceutical composition of claim 8, wherein said polyalkylene oxide is a monomethoxy-polyethylene glycol, (mPEG).

10. The pharmaceutical composition of claim 1, wherein said substantially non-antigenic polymer has a molecular weight of from about 200 to about 35,000.

11. The pharmaceutical composition of claim 10, wherein said substantially non-antigenic polymer has a molecular weight of from about 1,000 to about 15,000.

Claims 8 to 10 cover peginterferon alfa-2b in that the polyalkylene oxide is monomethoxypolyethylene glycol having an average molecular weight of 12,000-13,000 daltons. See Exhibit II Section 4.A.1 at pages 4 and 7.

Claim 13 of U.S. Patent No. 5,951,974 is:

13. A pharmaceutical composition, comprising a mixture of alpha interferon polymer conjugate positional isomers, wherein one of said positional isomers comprises an alpha interferon covalently conjugated to a substantially non-antigenic polymer at a histidine residue on said alpha interferon, wherein said substantially non-antigenic polymer is selected from the group consisting of polypropylene glycol, dextran, polyvinyl pyrrolidones, polyacryl amides, polyvinyl alcohols and carbohydrate-based polymers.

Claim 13 covers peginterferon alfa-2b product in that the product is a mixture of alfa interferon polymer conjugate positional isomers, wherein one of said positional isomers comprises an non-antigenic polymer covalently bonded to the alfa interferon at a histidine residue and wherein the non-antigenic polymer is polypropylene glycol. See discussions regarding Claims 1, and 8-10.

Claims 14-17 of U.S. Patent No. 5,951,974 are:

14. An alpha interferon-containing composition, comprising a plurality of alpha interferon polymer conjugates, wherein at least about 15% of the conjugates include covalent attachment of a substantially non-antigenic polymer at a histidine of said alpha interferon, wherein said substantially non-antigenic polymer is a polyalkylene oxide comprising an alkyl terminal.

15. The composition of claim 14, wherein the alpha interferon portion of said composition is alpha interferon 2b and said histidine is His34.

16. The composition of claim 14, wherein at least about 30% of said conjugates include covalent attachment of said substantially non-antigenic polymer at histidine-34 of said alpha interferon.

17. The composition of claim 16, wherein at least about 40% of said conjugates include covalent attachment of said substantially non-antigenic polymer at histidine-34 of said alpha interferon.

Claims 14-17 cover the peginterferon alpha 2b product in that approximately 47.8% of the conjugates include covalent attachment of the substantially non-antigenic polymer at histidine 34 of said alfa interferon. See Exhibit II, especially the Table on page 95.

Claim 18 of U.S. Patent No. 5,951,974 is:

18. A pharmaceutical composition, comprising a mixture of alpha interferon 2b-polymer positional isomers, wherein from about 30 to about 60% of the positional isomers include a substantially non-antigenic polymer conjugated to the His34 of said alpha interferon, from about 7 to about 20% of the positional isomers including a substantially non-antigenic polymer conjugated to the Cys1 of said alpha interferon and about 7 to about 15% of the positional isomers include a substantially non-antigenic polymer conjugated to the Lys121 of said alpha interferon, wherein said substantially non-antigenic polymer is a polyalkylene oxide comprising an alkyl terminal.

Claim 18 covers the peginterferon alfa-2b product in that said product comprises a mixture of alfa interferon 2b-polymer positional isomers wherein:

1) about 47.8% are conjugated to His34 of said alfa interferon;
2) about 13.2% are conjugated to Cys^{1} of said alfa interferon;
3) about 7.3% are conjugated to Lys^{121} of said alfa interferon; and
4) the substantially non-antigenic polymer – the polyethylene oxide comprising an alkyl terminal - is monomethoxy polyethylene glycol.

See also Exhibit II especially the Table on page 95 and the discussion regarding claims 8 and 9.

Claims 34-36 of U.S. Patent No. 5,951,974 are:

34. A method of treating an interferon-susceptible condition in mammals, comprising administering an effective amount of a composition of claim 1.

35. A method of treating an interferon-susceptible condition in mammals, comprising administering an effective amount of a composition of claim 14.

36. A method of treating an interferon-susceptible condition in mammals, comprising administering an effective amount of a composition of claim 18.

Claims 34-36 cover use of peginterferon alfa-2b powder for injection to treat the interferon susceptible condition - chronic hepatitis C. See Exhibit VII – FDA approval letter and U.S. Patent No. 5,951,974 at Col. 10 lines 4-10, 21-31 and 49-54 and Exhibit XII, Indications and Usage on page 2.

Claim 38 of U.S. Patent No. 5,951,974 is:

38. The pharmaceutical composition of claim 13, wherein said alpha interferon is interferon alpha 2b.

Claim 38 covers peginterferon alfa-2b in that the alfa interferon is interferon alfa-2b. See the discussion regarding Claims 2 and 13.

Claim 39 of U.S. Patent No. 5,951,974 is:

39. The pharmaceutical composition of claim 38, wherein said histidine residue is His34.

Claim 39 covers peginterferon alfa-2b. See the discussion regarding Claims 38 and 3.

Claims 40-42 of U.S. Patent No. 5,951,974 are:

40. The pharmaceutical composition of claim 13, wherein said mixture of said alpha interferon positional isomers comprises at least about 3 positional isomers.

41. The pharmaceutical composition of claim 13, wherein said mixture of said alpha interferon positional isomers comprises at least about 6 positional isomers.

42. The pharmaceutical composition of claim 13, wherein said mixture of said alpha interferon positional isomers comprises at least about 8 positional isomers.

Claims 40-42 cover peginterferon alfa-2b for reasons stated in reference to claims 13 and 4-7.

Claim 43 of U.S. Patent No. 5,951,974 is:

43. The pharmaceutical composition of claim 38, wherein said mixture of positional isomers comprises a substantially non-antigenic polymer linked to said alpha interferon 2b, at an amino acid residue selected from the group consisting of Cys1, Lys31, His34, Lys49, Lys83, Lys121, Lys131 and Lys134.

Claim 43 covers peginterferon alfa-2b for reasons stated in reference to claims 38 and 7 as well as claims 2 and 13.

Claims 44 and 45 of U.S. Patent No. 5,951,974 are:

44. The pharmaceutical composition of claim 13, wherein said substantially non-antigenic polymer has a molecular weight of from about 200 to about 35,000.

45. The pharmaceutical composition of claim 13, wherein said substantially non-antigenic polymer has a molecular weight of from about 1,000 to about 15,000.

Claims 44 and 45 cover peginterferon alfa-2b for reasons stated in reference to claims 13 and 10 and 11.

Claims 47 and 48 of U.S. Patent No. 5.951,974 are:

47. The pharmaceutical composition of claim 1 wherein said polyalkylene oxide is terminated with a C_{1-4} alkyl.

48. The pharmaceutical composition of claim 18 wherein said polyalkylene oxide is terminated with a C_{1-4} alkyl.

Claims 47 and 48 cover the peginterferon alfa-2b product for reasons stated in reference to claims 1, 9, 13 and 18.

Thus, at least claims 1-11, 13-18, 34-36, 38-45 and 47 and 48 of U.S. Patent No. 5,951,974 cover peginterferon alfa-2b, the active ingredient in PEG-Intron™ Powder for Injection, the approved product and a method of using PEG-Intron™ Powder for injection for treatment of chronic hepatitis C injections in patients of at least 18 years of age with compensated liver disease.

(10) A STATEMENT BEGINNING ON A NEW PAGE, OF THE RELEVANT DATES AND INFORMATION PURSUANT TO 35 U.S.C. §156(g) IN ORDER TO ENABLE THE SECRETARY OF HEALTH AND HUMAN SERVICES OR THE SECRETARY OF AGRICULTURE, AS APPROPRIATE, TO DETERMINE THE APPLICABLE REGULATORY REVIEW PERIOD AS FOLLOWS:

(i) FOR A PATENT CLAIMING A NEW DRUG, ANTIBIOTIC, OR HUMAN BIOLOGICAL PRODUCT, THE EFFECTIVE DATE OF THE INVESTIGATIONAL NEW DRUG (IND) APPLICATION AND THE IND NUMBER; THE DATE ON WHICH A NEW DRUG APPLICATION (NDA) OR A PRODUCT LICENSE APPLICATION (PLA) WAS INITIALLY SUBMITTED AND THE NDA OR PLA NUMBER AND THE DATE ON WHICH THE NDA WAS APPROVED OR THE PRODUCT LICENSE ISSUED:

Schering Corporation (Schering) of Kenilworth, New Jersey is the authorized agent of Enzon by virtue of the appointment of agent (Exhibit I) to Schering. Enzon is the assignee of record of United States Patent No. 5,951,974 by virtue of the Assignments of (1) Myung-ok Park-Cho (Exhibit VIII), and (2) Carl W. Gilbert (Exhibit IX).

In furtherance of the need for an approved NDA, Schering, on June 2, 1997 submitted to the FDA, a "Notice of Claimed Investigational New Drug"

for pegylated interferon alfa-2b (SCH 54031) under Section 351 of the Public Health Service Act for the purpose of conducting clinical studies to support the approval of a subsequent BLA for the use of PEG-Intron™ (pegylated interferon alfa-2b) Powder for Injection. A copy of this Schering letter is attached as Exhibit IV. By a letter dated June 5, 1997, the FDA acknowledged receipt of the IND and assigned the BB-IND No. 7173. In a July 1, 1997 memo FDA placed clinical hold for use of PEG-Intron BB-IND and requested additional data prior to Schering initiating clinical studies under BB-IND 7173. Schering continued to work with FDA to provide such additional information, amendments, and data and on July 29, 1997 FDA removed clinical hold for BB-IND 7173. This establishes the beginning of the "regulatory review period" under 35 U.S.C. §156(g)(1)(B)(i) as July 29, 1997, the effective date of an investigational exemption.

Schering submitted an original Biologics License Application (BLA) for PEG-Intron™ (peginterferon alfa-2b) therapy for treatment of chronic hepatitis C in humans on December 22, 1999. A copy of this Schering letter transmitting the BLA is attached as Exhibit VI.

By a letter dated January 19, 2001 (copy attached as Exhibit VII), the FDA advised Schering that BL 103949 replaces Ref. BLA No. 99-1488 and that BLA 99-1488 was approved on January 19, 2001 for peginterferon alfa-2b for treatment of chronic hepatitis C in patients not previously treated who have compensated liver disease and are at least 18 years of age.

Thus, for purposes of determining the "testing phase" of the "regulatory review period" under 35 U.S.C. §156(g)(1)(B)(i), the "testing phase" began on July 29, 1997, the effective date of the BB-IND No. 7173 ended on December 22, 1999, the date the BLA No. 99-1488 was initially submitted by Schering for use of PEG-Intron™ (peginterferon alfa-2b) powder for injection for treatment of chronic hepatitis C in humans under §351 of the Public Health Service Act. And, for purposes of determining the "approval phase" of the "regulatory review period" under 35 U.S.C. §156(g)(1)(B)(ii) the "approval phase" began on December 22, 1999, the date the BLA No. 19-1488 was initially submitted by Schering to the FDA and ended on January 19, 2001, the date on which the BLA No. 99-1488 was approved by the FDA.

(11) A BRIEF DESCRIPTION BEGINNING ON A NEW PAGE OF THE ACTIVITIES UNDERTAKEN BY SCHERING, THE MARKETING APPLICANT, DURING THE APPLICABLE REGULATORY REVIEW PERIOD WITH RESPECT TO THE APPROVED PRODUCT AND THE SIGNIFICANT DATES APPLICABLE TO SUCH ACTIVITIES:

During the applicable regulatory review period, Schering was actively involved in obtaining FDA approval for the use of PEG-Intron™ (peginterferon alfa-2b) powder for injection for treatment of chronic hepatitis C in patients who have compensated liver disease and are at least 18 years of age. As previously noted, Schering submitted a BB-IND on June 2, 1997. On June 5, 1997, FDA acknowledged receipt of the Schering IND and assigned it number BB-IND 7173. On July 1, 1997, FDA placed a clinical hold on BB-IND 7173 and requested additional data prior to Schering initiating clinical studies under BB-IND 7173. During July 1997, Schering continued to work with FDA to supply additional data, amendments and information. On July 29, 1997, FDA removed the clinical hold on BB-IND 7173. During the period July 29, 1997 to December 22, 1999, Schering in close consultation with FDA, conducted clinical trials under BB-IND 7173. Schering submitted on December 22, 1999, BLA No. 99-1488 for use of PEG-Intron™ (peginterferon alfa-2b) in the treatment of chronic hepatitis C in humans. From December 22, 1999 to January 19, 2001, Schering continued to interact with various FDA officials and answered numerous questions, generated requested data and supplied requested information regarding all clinical studies and data on the use of PEG-Intron™ (peginterferon alfa-2b)

for the treatment of chronic hepatitis C in patients not previously treated with interferon alfa who have compensated liver disease. A brief description of the significant activities undertaken by Schering with respect to the use of PEG-Intron™ (peginterferon alfa-2b) powder for injection for the treatment of chronic hepatitis C during the regulatory review period is set forth in Exhibit X (BB-IND) and Exhibit XI (BLA) and is illustrative of the activities involved.

(12) A STATEMENT BEGINNING ON A NEW PARAGRAPH THAT IN THE OPINION OF THE APPLICANT THE PATENT IS ELIGIBLE FOR AN EXTENSION AND A STATEMENT AS TO THE LENGTH OF THE EXTENSION CLAIMED, INCLUDING HOW THE LENGTH OF EXTENSION WAS DETERMINED:

(a) Statement of eligibility of the patent for extension under 35 U.S.C. §156(a):

Section 156(a) provides, in the relevant part, that the term of a patent which claims a product, a method of using a product, or a method of manufacturing a product shall be extended if (1) the term of the patent has not expired before an application for extension is submitted; (2) the term of the patent has never been extended under 35 U.S.C. §156(e)(1); (3) the application for extension is submitted by the owner of record of the patent or its agent in accordance with 35 U.S.C. §156(d); (4) the product has been subject to a regulatory review period before its commercial marketing or use; and (5) the permission for the commercial marketing or use of the product after such regulatory review period is the first permitted commercial marketing

or use of the product using the provision of law under which such regulatory review period occurred.

As described below by corresponding number, each of these elements is satisfied here:

(1) Pursuant to 35 U.S.C. §154(c)(1), as amended (effective June 8, 1995) by the Uruguay Round Agreements Act, Publ. 103-465, 108 Stat. 4809 (1994) and 35 U.S.C. §156, the term of United States Patent No. 5,951,974 currently expires on November 10, 2013. This application is, therefore, being submitted prior to the expiration of the term of United States Patent No. 5,951,974.

(2) The term of this patent has never been extended under 35 U.S.C. §156(e)(1).

(3) This application is being submitted by Schering Corporation, by virtue of the appointment of agent to Schering Corporation by Enzon, the owner of record of this patent (Exhibit I). Enzon is the owner of record by virtue of the Assignments of (1) Myung-ok Park-Cho (Exhibit VIII) and (2) Carl W. Gilbert (Exhibit IX). This application is submitted in accordance with 35 U.S.C. §156(d) in that it is submitted within the sixty-day period beginning on January 19, 2001, the date the product received permission for marketing under Section 351 of the Public Health Service Act and ending on January 19, 2001 and contains the information required under 35 U.S.C. §156(d).

(4) As evidenced by the January 19, 2001 letter from the FDA (Exhibit VII), to Schering Corporation, the product was subject to a regulatory review period under Section 351 of the Public Health Service Act before its commercial marketing or use.

(5) Finally, the PEG-Intron™ (peginterferon alfa-2b) powder for injection product was approved by the FDA for treatment of adult patients with chronic hepatitis C. The permission for the commercial marketing of PEG-Intron™ (peginterferon alfa-2b) powder for injection after regulatory review under Section 351 of the Public Health Service Act [42 U.S.C. §262(a)(2)(B)], is the first permitted commercial marketing and use under Section 351 for humans of the active ingredient peginterferon in PEG-Intron™ (peginterferon alfa-2b) powder for injection. This is confirmed by the absence of any approved new drug application for the active ingredient in humans prior to January 19, 2001. See Exhibit VII.

(b) Statement as to length of extension claimed:

The 17 year from filing term of United States Patent No. 5,951,974 now expiring on November 10, 2013 should be extended by 435 days to January 19, 2015. This extension was determined on the following basis. As set forth in 35 U.S.C. §156(g)(1), the regulatory review period equals the length of time between the effective date of the BB-IND No. 7173 of July 29, 1997 and the submission of the BLA on December 22, 1999, a period of 876 days, plus the length of time between the submission of the BLA on

December 22, 1999 to BLA approval on January 19, 2001, a period of 394 days. These two periods added together equal 1,270 days.

Pursuant to the introduction of 35 U.S.C. §156(c), the term of the patent eligible for extension shall be extended only for that portion of the regulatory review period which occurs after the date the patent is issued. In this case, the limitation under the introduction to §156(c) applies in that the issue date of United States Patent No. 5,951,974 (September 14, 1999) is after the date on which the regulatory review period began. Thus, the period calculated under §156(g)(1)(B)(i) is reduced to 99 days.

Section 156(c)(2), requires the period calculated under §156(g)(1)(B)(i) to be reduced by one-half of the 99 day period; this reduction results in a value of 50 days.

From the foregoing calculation, an extension of 444 days results, i.e., the period under 35 U.S.C. 156(g)(1`)(B)(i) as limited by §156(c)(2) (50 days) plus the period under 35 U.S.C. 156(g)(1)(B)(ii) (394 days). This extension period is subject to two further potential limitations under §156.

First, under §156(g)(6)(A), a maximum extension of five years is permitted. Since the calculated extension (444 days) is less than five years (1,827 days), this limitation does not apply (the patent issued on September 14, 1999, which was after the enactment of §156 in 1984).

Second, under §156(c)(3), if the period remaining in the term of the patent after the date of approval, that is, January 19, 2001 to November 10, 2013, when added to the extension period calculated above would exceed 14 years, the period of extension would be limited so that the total does not exceed 14 years. In this case, however, the total of the remaining term (4,678 days) plus the 444 day extension is 5,122 days and does exceed the 14 year (5,113 days) limit, and the extension is thereby reduced to 435 days.

Accordingly, United States Patent No. 5,951,974 is eligible for a 435 day extension from November 10, 2013 to January 19, 2015.

(13) A STATEMENT ON A NEW PAGE THAT APPLICANT ACKNOWLEDGES A DUTY TO DISCLOSE TO THE COMMISSIONER OF PATENTS AND TRADEMARKS AND THE SECRETARY OF HEALTH AND HUMAN SERVICES ANY INFORMATION WHICH IS MATERIAL TO THE DETERMINATION OF ENTITLEMENT TO THE EXTENSION SOUGHT.

Schering acknowledges a duty to disclose to the Commissioner of Patents and Trademarks and the Secretary of Health and Human Services any information which is material to the determination of entitlement to the extension sought.

As stated in Paragraph No. 9 hereinabove Schering asserts that at least claims 1-11, 13-18, 38-45 and 47 and 48 of U.S. Patent No. 5,951,974 cover peginterferon alfa-2b in the approved product PEG-Intron™ (peginterferon alfa-2b) powder for injection.

The term of United States Patent No. 5,591,974 has never been extended. A copy of this patent is attached as Exhibit III.

(14) PRESCRIBED FEES:

The Commissioner is authorized to charge our Deposit Account No. 19-0365 in the amount of $1,120.00 or any other fee necessary for this application to prevent it from becoming inadvertently abandoned.

(15) THE NAME, ADDRESS AND TELEPHONE NUMBER OF THE PERSON TO WHOM INQUIRIES AND CORRESPONDENCE RELATING TO THIS APPLICATION FOR PATENT TERM EXTENSION ARE TO BE DIRECTED TO:

THOMAS D. HOFFMAN
SCHERING-PLOUGH CORPORATION
PATENT DEPARTMENT (K-6-1- 1990)
2000 GALLOPING HILL ROAD
KENILWORTH, NEW JERSEY 07033-0530
TEL. NO. (908) 298-5037
FACSIMILE NO. (908) 298-5388

(16) CERTIFICATION THAT THE ENCLOSED DUPLICATE COPY OF THIS APPLICATION IS A TRUE COPY OF THE ORIGINAL:

I, Thomas D. Hoffman, Registration No. 28,221, as duly appointed attorney (by virtue of the following Power of Attorney duly executed by James R. Nelson, Vice President for Schering Corporation) for Applicant, Schering Corporation, authorized agent (by virtue of the Appointment of Agent, see Exhibit I) for the owner of record of United States Patent No. 5,951,974 (by virtue of the aforesaid Assignments, see Exhibits (VIII & IX) which has applied for an extension of term of this patent, declare that the duplicate copy of this application transmitted herewith is a true copy of the original application.

I hereby acknowledge that all statements made herein of my own knowledge are true and that all statements made on information or belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of this application and any extension of United States Patent No. 5,951,974.

Date: March 4 2001

Thomas D. Hoffman
Attorney for Authorized Agent
of the Assignee of Record
Registration No. 28221
Tel. No. (908) 298-5037

DECLARATION AND POWER OF ATTORNEY
BY AUTHORIZED AGENT

As the below identified official of Schering Corporation, the authorized agent for the owner of record of United States Patent No. 5,951,974, which has applied for an extension of term of this patent, I declare (1) that I have been authorized to practice before the United States Patent and Trademark Office; and (2) that I have general authority from Schering Corporation, the authorized agent of the owner of record, to act on behalf of the owner of record in patent matters.

I hereby acknowledge that all statements made herein of my own knowledge are true and that all statements made on information or belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of this application and United States Patent No. 5,951,974.

POWER OF ATTORNEY: I hereby appoint as United States attorneys and with full powers of substitution and revocation, to prosecute this application and transact all business in the Patent and Trademark Office connected therewith: Thomas D. Hoffman, Reg. No. 28,221; Henry S. Hadad, Reg. No. 35,888; Anita W. Magatti, Reg. No. 29,825; Edward H. Mazer, Reg. No. 27,573; Robert A. Franks, Reg. No. 29,605 and Richard J. Grochala, Reg. No. 31,518.

Send correspondence to:

Thomas D. Hoffman
Schering-Plough Corporation
Patent Dept., K-6-1-1990
2000 Galloping Hill Road
Kenilworth, NJ 07033-0530
Tel. No. (908) 298-5037

Date: March 1, 2001

By: James R. Nelson
Vice President,
Schering Corporation
Reg. No. 27,929

(17) DECLARATION FOR EXTENSION OF UNITED STATES PATENT NO. 5,591,974

I, THOMAS D. HOFFMAN, Registration No. 28,221, as duly appointed attorney (by virtue of the Power of Attorney duly executed by James R. Nelson, Vice President for Schering Corporation) for Applicant, Schering Corporation, the authorized agent for CRCT (by virtue of the Power of Appointment, see Exhibit I), the owner of record of United States Patent No. 5,951,974 (by virtue of the aforesaid Assignments, see Exhibits VIII & IX) which has applied for an extension of term of this patent, declare that

(1) I have been authorized to practice before the United States Patent and Trademark Office;

(2) I have reviewed and understand the contents of the attached application for extension of United States Patent No. 5,951,974;

(3) I believe that the patent is subject to extension under 35 U.S.C. §156 and 37 C.F.R. §1.710;

(4) I believe that the length of extension claimed for the 17 year from filing date term specified in paragraph 12 is fully justified pursuant to 35 U.S.C. §156 and the applicable regulations; and

(5) I believe that the patent for which an extension is being sought meets the conditions for extension of the term of a patent as set forth in 35 U.S.C. §156 and 37 C.F.R. §1.720.

I hereby acknowledge that all statements made herein of my own knowledge are true and that all statements made on information or belief are believed to be true; and further that these statements were made with the

knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of this application and any extension of United States Patent No. 5,951,974.

Date: March 4 2001 Thomas D Hoffman

Thomas D. Hoffman
Attorney for Authorized Agent
for the Assignee of Record
Reg. No. 28,221
Tel. No. (908) 298-5037

EXHIBIT I

APPOINTMENT OF AGENT

WHEREAS, ENZON, INC. (hereinafter "ENZON"), a company organized and existing under the laws of the State of Delaware, U.S.A., and having its principal office at 20 Kingsbridge Road, Piscataway, New Jersey 08854-3998, is the owner of record of U.S. Patent No. 5,591,974, entitled, "INTERFERON POLYMER CONJUGATES," which was granted on September 14, 1999, by virtue of assignments of such U.S. patent to ENZON, recorded in the United States Patent and Trademark Office at Reel 9063, Frame 0929 on December 22, 1997, and at Reel 9063, Frame 0961 on March 6, 1998;

WHEREAS, SCHERING CORPORATION (hereinafter "SCHERING"), a corporation organized and existing under the laws of the State of New Jersey, U.S.A., with its principal offices at 2000 Galloping Hill Road, Kenilworth, New Jersey 07033, has entered into an agreement with ENZON under which SCHERING was granted certain rights under U.S. Patent No. 5,951,974;

WHEREAS, SCHERING is desirous of marketing a product containing a composition within the scope of the claims of U.S. Patent No. 5,951,974 including the product known as PEG-INTRON (peginterferon alfa-2b) Powder for Injection;

WHEREAS, SCHERING received marketing approval on January 19, 2001 from the United States Food and Drug Administration to market PEG-INTRON (peginterferon alfa-2b) Powder for Injection; and

WHEREAS, 35 U.S.C. § 156, entitled, "Extension of Patent Term," provides at Section (a)(3) that an application for extension of a patent term be submitted by the owner of record or its agent.

NOW, THEREFORE, as the below-identified official of ENZON, I state that (1) I have been authorized to obligate ENZON to sign this Appointment of Agent; and (2) I hereby appoint SCHERING, its subsidiaries and/or its designees as agents of ENZON for the express purpose of submitting and handling all matters and correspondence in the U.S. Patent and Trademark Office attendant to the application for extension of the term of U.S. Patent No.5,951,974 covering PEG-INTRON(peginterferon alfa-2b) Powder for injection pursuant to 35 U.S.C. § 156. This appointment shall be co-extensive with the term of the aforesaid agreement between ENZON and SCHERING.

ENZON, INC.

Date: February 26, 2001

By: _____
Name: Peter G. TOMBROS
Title: President and Chief Executive Officer

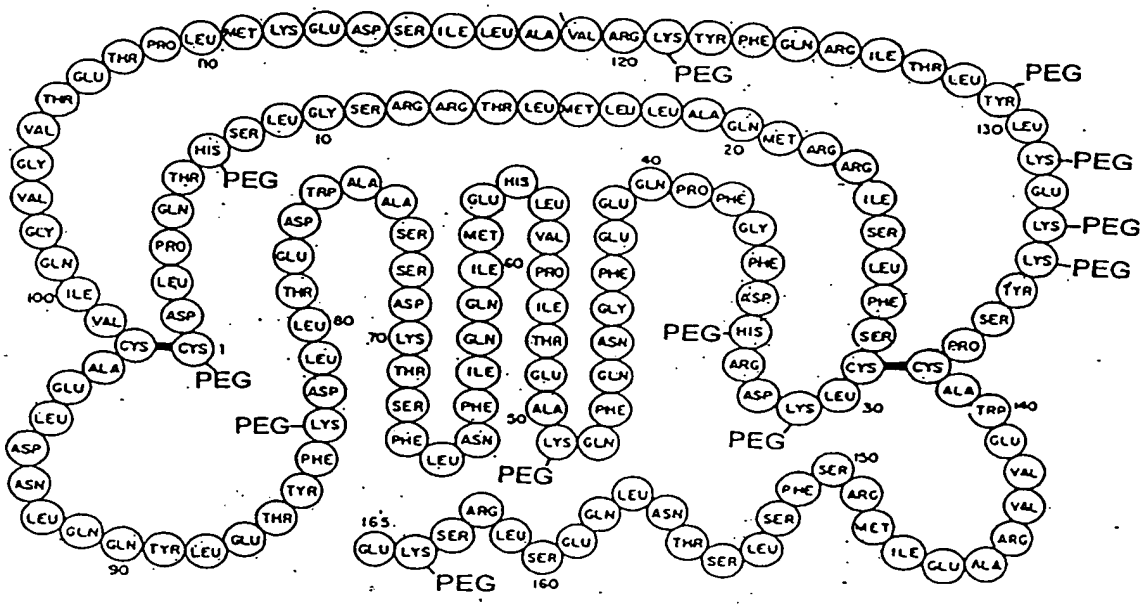
EXHIBIT II

4.A.1.1 Description

Physical Description:

Clear to opalescent, colourless to slightly yellow solution; essentially free of visible particles.

Chemical Structure:



Note: The predominant form is one PEG molecule attached to each IFN molecule; all positional isomers of the pegylated product have been identified and include attachment of one PEG molecule at any of 12 different sites including 8 Lysines, 2 Histidines, 1 Tyrosine, and N-terminal Cysteine.

Molecular Weight:

Approximately 31,870 daltons (due to PEG polymer heterogeneity)

USAN name: peginterferon alfa-2b

Code Name: SCH 54031

CAS Number: 215647-85-1

Other Names: PEG-IFN

PEG-Interferon alfa-2b

PEG-Intron (commercial product)

1. INTRODUCTION

SCH 54031 PEG_{12000}-Interferon alfa-2b (PEG-IFN) is a covalent conjugate of recombinant Interferon alfa-2b (IFN) with monomethoxypolyethylene glycol (PEG, average molecular weight of 12,000 daltons). PEG-IFN is synthesized by the reaction of IFN drug substance with an electrophilic derivative of PEG, succinimidylcarbonate PEG (SC-PEG), in 100 mM sodium phosphate (pH 6.5). Numerous nucleophilic sites on IFN exist, including the ε-amino groups of the 10 Lysines, the α-amino group of the N-terminal Cysteine, the imidazolyl nitrogens of the 3 Histidines, and the hydroxyl groups of the 14 Serines, 10 Threonines, and 5 Tyrosines. As shown in Figure 1, SC-PEG can theoretically react with amino groups to yield PEG-carbamate derivatives, with Histidyl imidazole groups to produce PEG-oxycarbonyl imidazole derivatives, and with hydroxy groups to give PEG-carbonate derivatives of IFN. Various factors, including solvent accessibility, protein conformation, and local electronic and pK effects determine the ultimate reactivity of the sites.

Following the reaction, the material is diafiltered and loaded onto an anion-exchange column and eluted using a salt gradient. The pooled column fractions are concentrated and diafiltered to yield the final purified bulk solution. Figure 2 provides a summary of the production and purification scheme of PEG-IFN. The resulting product is a heterogeneous population of molecules, consisting mainly of mono-pegylated IFN with smaller amounts of dipegylated and non-pegylated IFN. Detailed characterization of PEG-IFN is presented in this report.

2. STRUCTURE VALIDATION

2.1. Mass Spectrometric Analysis

Matrix-Assisted Laser Desorption Ionization Mass Spectrometry (MALDI-MS) was used to characterize PEG-IFN (see Figure 3). The matrix used was sinapinic acid and analysis was performed on a Voyager Elite Mass Spectrometer (PerSeptive Biosystems) using time-of-flight analysis at The University of Texas at Houston Health Sciences Center. The MALDI-MS spectra obtained for PEG-IFN shows peaks corresponding to the $[M + H]^{+1}$, $[M + 2H]^{+2}$, and $[2M + H]^{+1}$ ions of mono-pegylated IFN at masses 31821.2, 16033.5, and 63744.4, respectively. The broadness of the peaks in the PEG-IFN mass spectra is due to the heterogeneous nature of the "PEG 12,000" which is a mixture of polyethylene glycol polymers having a distribution of molecular weights with an average mass in the 12,000-13,000 Da mass range. The ion with mass 19278.2 is due to the $[M + H]^{+}$ ion of free IFN, which is present in the sample in low amounts and which is also generated to limited extents by laser irradiation of pegylated IFN. The difference in mass of IFN and the major pegylated species in the mass spectrum is 12543 Da, which is approximately the average mass of one PEG group. Therefore, the mass spectra of the predominant ion in PEG-IFN is consistent with the presence of one PEG group bound to IFN.

2.2. Tryptic Digest Mapping

Both PEG-IFN and IFN were digested by incubation with trypsin at 37°C for 18 hours in sodium phosphate buffer, pH 7.0. Aliquots of each digest were analyzed by HPLC using a Delta Pak C18 column, 5 µm particle size, 150 x 3.9 mm (Waters), 300A pore size. A gradient method employing mobile phases of 0.1% TFA in water and 90/10 acetonitrile/aqueous 1% TFA was used with detection at 214 nm. The

the 15 fractions (see Table 4) demonstrate consistent batch-to-batch reproducibility for the positional isomer profile.

Table 4 Area Percentages of Positional Isomers of PEG-IFN as Separated by Cation Exchange Chromatography

	Peak Numbers														
Ba.#	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
7-PPI-101	3.3	5.9	1.3	1.4	1.2	47.9	0.9	6.9	7.3	3.4	0.8	4.3	1.2	13.5	0.8
7-PPI-102	3.2	5.6	1.2	1.7	1.1	47.6	0.8	6.8	7.1	3.4	0.8	4.6	1.2	14.1	0.9
7-PPI-103	3.4	5.7	1.2	1.5	1.1	48.0	0.9	6.9	7.1	3.6	0.8	4.6	1.1	13.7	0.7
7-PPI-104	3.9	6.0	1.2	1.4	1.1	47.5	0.9	6.7	6.9	3.4	0.8	4.3	1.2	13.8	0.8
7-PPI-105	3.9	5.5	1.2	1.7	1.4	46.9	0.9	6.7	6.9	3.5	0.7	4.5	1.2	14.5	0.7
6-PPI-101	3.6	5.6	1.3	1.4	1.1	47.6	0.8	7.2	7.9	4.0	0.8	4.5	1.2	12.2	0.9
6-PPI-102	3.5	5.7	1.4	1.3	1.1	48.1	0.8	7.0	7.6	3.8	0.9	4.8	1.0	12.4	0.7
6-PPI-103	3.7	5.4	1.5	1.7	1.3	47.9	0.8	7.0	7.2	3.6	1.1	4.7	0.9	12.6	0.6
6-PPI-104	3.5	5.7	1.4	1.3	1.2	49.3	0.3	6.6	7.6	3.6	1.1	4.6	0.9	12.3	0.6
Avg	3.6	5.7	1.3	1.5	1.	47.8	0.8	6.9	7.3	3.6	0.9	4.5	1.1	13.2	0.7

2.7.2. Isolation of Positional Isomers via Preparative Cation Exchange Chromatography

Preparative isolations of PEG-IFN positional isomers were performed using the same type of TosoHaas cation exchange column as that used for the analytical scale fractionation of positional isomers but having larger dimensions (21.5 mm x 15 cm). Mobile phases were the same as those used for the analytical separations, but the flow rates were increased and the gradient adjusted to accommodate the larger column and larger amounts of protein injected onto the column. Various batches of PEG-IFN Drug Substance were pooled, including batches 7-PPI-101, 7-PPI-102, 7-PPI-103, 7-PPI-104, 7-PPI-105, and 35953-011, and concentrated approximately 10-fold and buffer exchanged into mobile phase A using Centricon 10 microconcentrators, having 10 kD cutoff filters. Using injection volumes of 200 uL and detection at 214 nm, fractions were collected corresponding to the peaks shown in Figure 21. The collected fractions were concentrated with Centricon 10

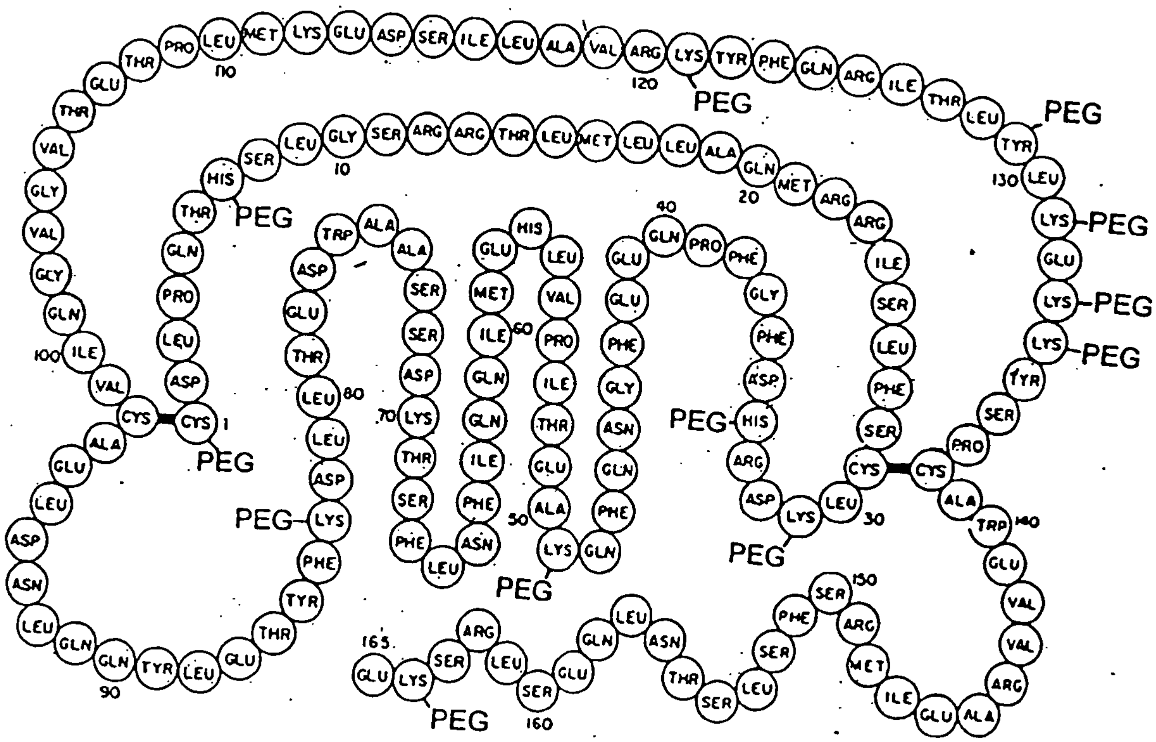


Figure 50 Sites of Pegylation in PEG-IFN

Note: Only one site is pegylated in any one positional isomer.

3. CONCLUSION

Structure validation studies were performed on PEG-IFN. The basic sequence of IFN appears to be preserved in PEG-IFN as evidenced by the similarity of tryptic digests of IFN and PEG-IFN when they are analyzed by RP-HPLC and MALDI-MS. Also, immunoblot analysis of PEG-IFN indicates that the epitope of IFN that binds the NK-2 monoclonal antibody is preserved in PEG-IFN. Mass spectrometric, HPSEC, and SDS-PAGE analyses of PEG-IFN are consistent with mono-pegylated IFN as the major component of PEG_{12000}-IFN. Dipegylated and non-pegylated IFN are minor components. Characterization studies of PEG-IFN indicate the following positions on IFN are derivatized by PEG (also referred to as positional isomers, see Figure 50):

Amino Acid Position	Approximate Abundance of Pegylated Species*
His^{34}	47.8%
Cys^{1}	13.2%
Lys^{121}	7.3%
Lys^{31}	5.7%
Lys^{49}	4.5%
Lys^{83}	3.6%
Lys^{131}	3.5%
Lys^{164}	3.4%
Lys^{133}	1.5%
Lys^{134}	<1.3%
His^{7}	0.9%
Tyr^{129}	0.8%

* Data represent the mean values obtained from analysis of nine batches of PEG-IFN by HPIEC (see Table 4)

Comparison of the CD spectra of PEG-IFN and IFN indicate no significant secondary/tertiary conformational differences exist between these materials. Finally, PEG-IFN when assayed as a mixture of di-PEG-IFN, mono-PEG-IFN, and non-pegylated IFN species has approximately 25% of native IFN antiviral bioactivity. After isolation by HPSEC, the di-pegylated component had ~3% of native IFN activity. The mono-pegylated components had approximately 20% of native IFN activity. The positional isomers of monopegylated IFN differ in both the stability of PEG attachment as well as bioactivities. The major positional isomer, pegylated at His^{34}, has a relatively high bioactivity and moderate stability.

US005951974A

United States Patent [19]

Gilbert et al.

[11] Patent Number: 5,951,974

[45] Date of Patent: Sep. 14, 1999

EXHIBIT III

[54] INTERFERON POLYMER CONJUGATES

[75] Inventors: **Carl W. Gilbert**, Powder Springs, Ga.; **Myung-ok Park-Cho**, Seoul, Rep. of Korea

[73] Assignee: **Enzon, Inc.**, Piscataway, N.J.

[21] Appl. No.: **08/994,622**

[22] Filed: **Dec. 19, 1997**

Related U.S. Application Data

[63] Continuation-in-part of application No. 08/337,567, Nov. 10, 1994, Pat. No. 5,711,944, which is a continuation-in-part of application No. 08/150,643, Nov. 10, 1993, abandoned.

[51] **Int. Cl.**[6] **A61K 38/21**; C07K 1/113; C07K 14/56

[52] **U.S. Cl.** **424/85.7**; 530/351; 530/409; 530/410; 530/411

[58] **Field of Search** 424/85.4, 85.7; 530/351, 409, 410, 411

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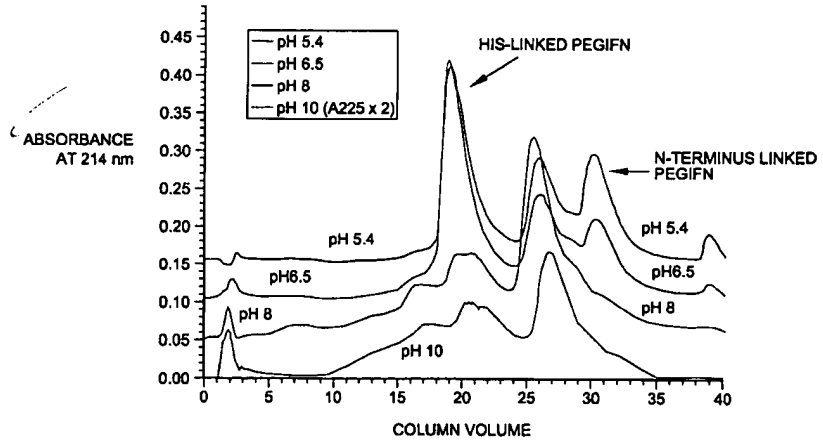
Primary Examiner—Jeffrey E. Russel
Attorney, Agent, or Firm—Roberts & Mercanti, LLP

[57] ABSTRACT

Compositions containing alpha interferon conjugated to a substantially non-antigenic polymer are disclosed in which at least about 30% of the conjugates include covalent attachment of the alpha interferon to the substantially non-antigenic polymer at a histidine. Also disclosed is a process for preparing the conjugates. The process includes contacting an alpha interferon with a succinimidyl carbonate-activated substantially non-antigenic polymer at a pH which is sufficient to facilitate covalent attachment of the polymer on a histidine of the alpha interferon.

50 Claims, 2 Drawing Sheets



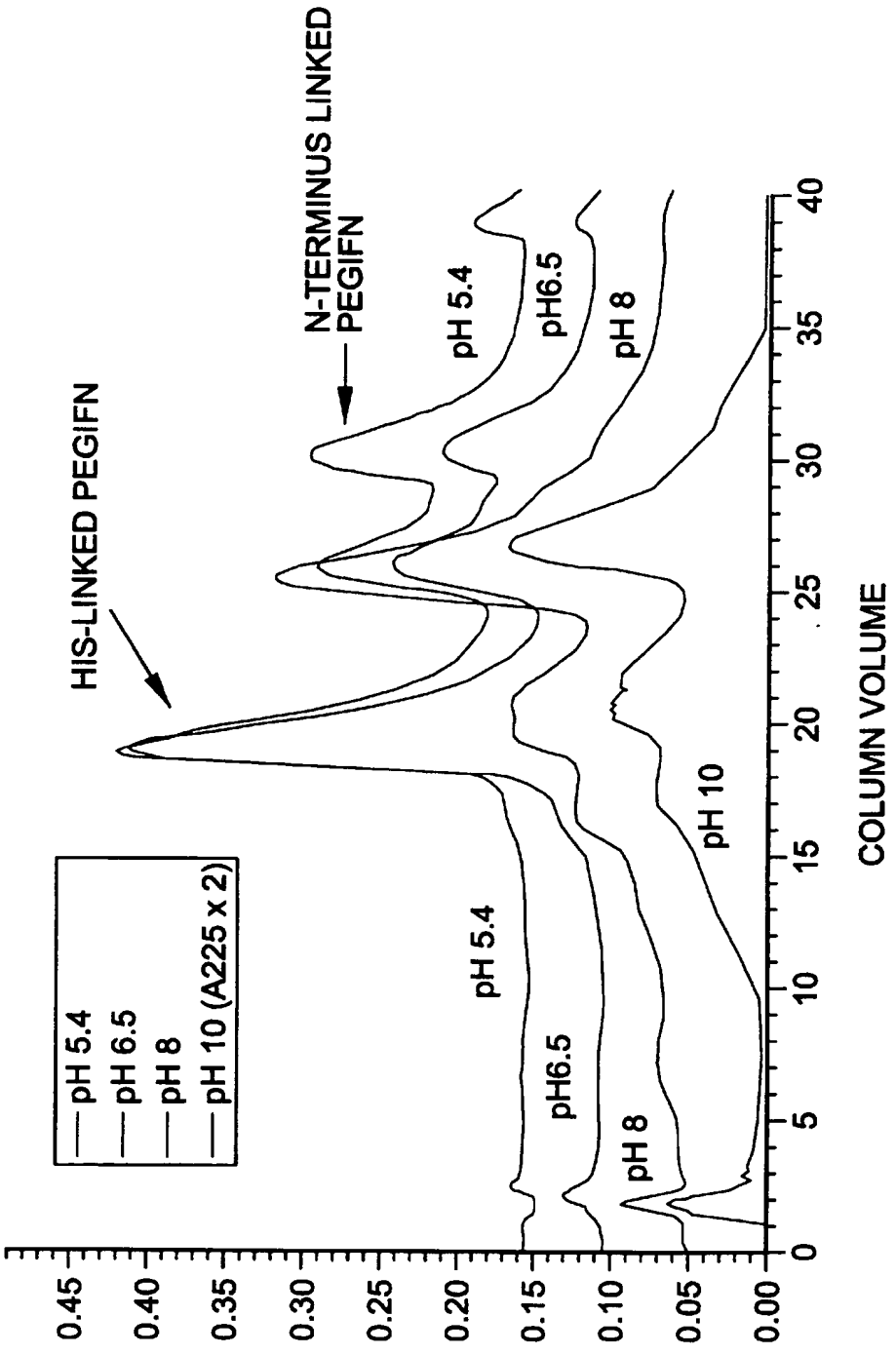
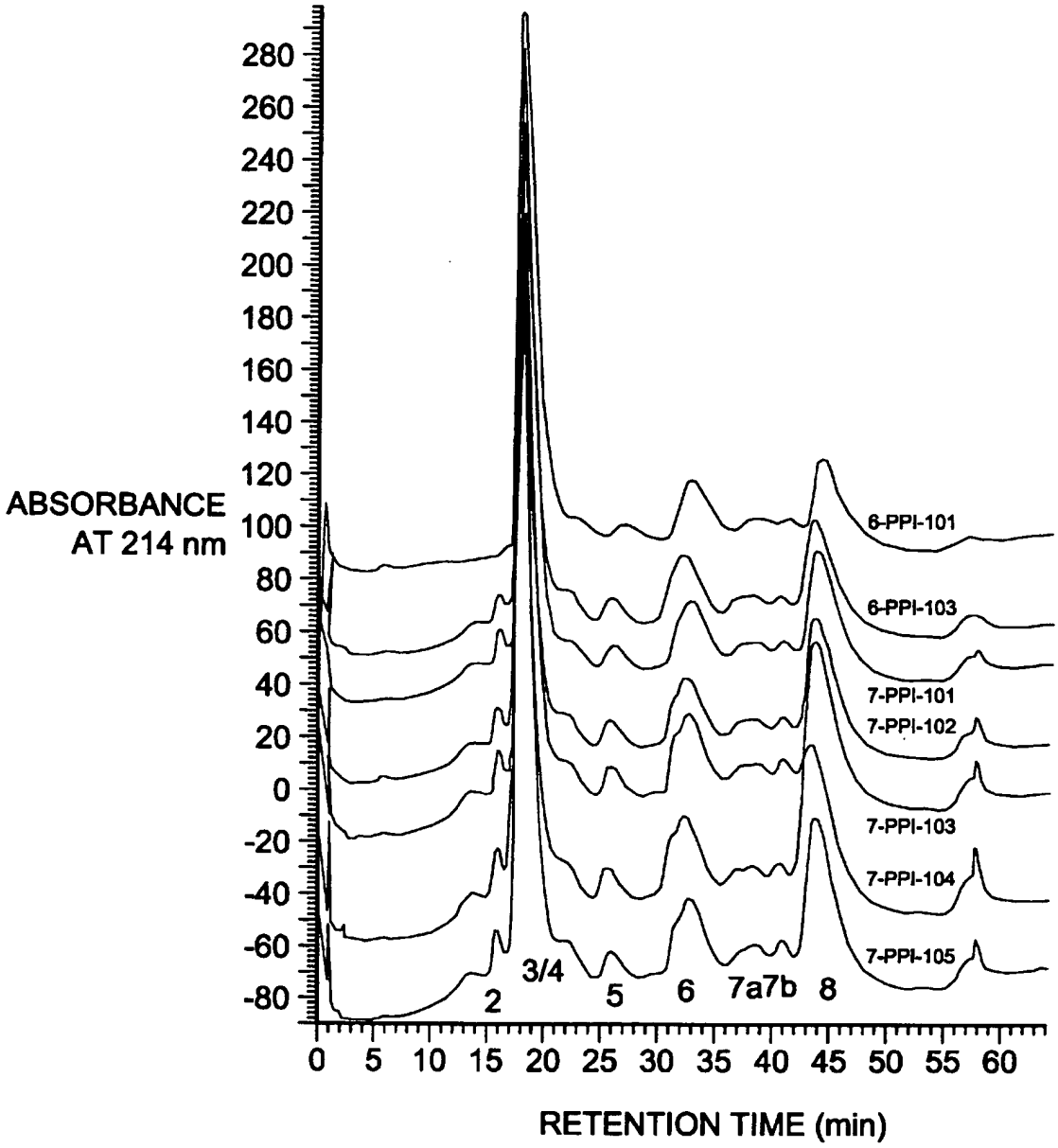

HIS-LINKED PEGIFN
N-TERMINUS LINKED PEGIFN
pH 5.4
pH6.5
pH 8
pH 10
pH 10 (A225 x 2)
ABSORBANCE AT 214 nm
COLUMN VOLUME
0.00
0.05
0.10
0.15
0.20
0.25
0.30
0.35
0.40
0.45
0
5
10
15
20
25
30
35
40

FIG-1

FIG-2


ABSORBANCE
AT 214 nm
280
260
240
220
200
180
160
140
120
100
80
60
40
20
0
-20
-40
-60
-80
6-PPI-101
6-PPI-103
7-PPI-101
7-PPI-102
7-PPI-103
7-PPI-104
7-PPI-105
2
3/4
5
6
7a7b
8
0 5 10 15 20 25 30 35 40 45 50 55 60
RETENTION TIME (min)

INTERFERON POLYMER CONJUGATES

This application is a continuation-in-part of U.S. patent application Ser. No. 08/337,567, filed Nov. 10, 1994, now U.S. Pat. No. 5,711,944 which, in turn, is a continuation-in-part of U.S. patent application Ser. No. 08/150,643, filed Nov. 10, 1993, now abandoned. The contents of each application are incorporated herein by reference.

BACKGROUND OF THE INVENTION

1. Field of the Invention

The present invention is directed to interferon-polymer conjugates. In particular, the invention is directed to conjugates having a novel interferon-polymer attachment profile.

2. Description of Related Art

Conjugating biologically-active proteins to polymers has been suggested to improve one or more of the properties of circulating life, water solubility or antigenicity in vivo. For example, some of the initial concepts of coupling peptides or polypeptides to polyethylene glycol (PEG) and similar water-soluble polymers are disclosed in U.S. Pat. No. 4,179,337, the disclosure of which is incorporated herein by reference.

Insulin and hemoglobin were among the first therapeutic agents conjugated. These relatively large polypeptides contain several free lysine ε-amino attachment sites. Several polymers could be attached without significant loss of biologic activity.

For many biologically active materials, the conjugation process, however, is not without complications. Care must be taken to limit the loss of biological activity caused by the conjugation reaction. For example, if too much of the activated polymer is attached to the target protein or polypeptide, biological activity can be severely reduced or lost. Further, if the wrong linker joining the polymer to the protein is used or an insufficient amount of polymer is attached to the target, the therapeutic value of the resultant conjugate is rather limited. Often, such conjugates do not demonstrate enough of an increase in the circulating life to compensate for the loss in bioactivity. Problems can also result when a therapeutic moiety's active site (i.e. where groups associated with bioactivity are found) becomes blocked as a result of the polymer attachment. This problem can be difficult to avoid since the polymer and protein are typically joined in solution-based reactions. Pre-blocking the active sites with materials such as pyridoxal phosphate has been suggested, but the results have been inconsistent. The problems are particularly acute with lower molecular weight proteins and peptides. These bioactive materials often have few attachment sites not associated with bioactivity.

Interferons, hereinafter also referred to as IFN's, are a particular example of proteins which could benefit from improved polymer conjugation techniques. See, for example, U.S. Pat. Nos. 4,766,106 and 4,917,888 which describe inter alia beta interferon conjugated with activated polymers including mPEG-2,4,6-trichloro-S-triazine, mPEG-N-succinimidyl glutarate or mPEG-N-succinimidyl succinate. The patentees disclose that covalent modification of the protein is done at a pH of from 5 to 9 and, when the protein is reacted through its lysine residues, covalent modification of the protein is done at a pH of from 8 to 9. Relatively high molar excesses (10, 20 and 50-fold) of the activated polymer are also used.

European Patent Application bearing publication No. 0 236 987 describes reacting alpha and gamma interferons with high molar excesses of alkyl imido ester-activated polyethylene glycols under conditions which preferably include a pH of from approximately 7 to 9. European Patent Application bearing publication No. 0 510 356 describes conjugating alpha interferon with pyridinyl carbonyl and thiocarbonyl activated PEG at a pH of from 7 to 9. There was no mention in these disclosures that amino acids other than lysine were involved in the conjugation or that it would be advantageous to do so.

In spite of the above-described disclosures, most interferon-polymer conjugates have been deemed to be unacceptable for one reason or another.

The present invention addresses these shortcomings.

SUMMARY OF THE INVENTION

In one aspect, the present invention includes pharmaceutical compositions containing a mixture of mono-polymer stranded alpha interferon conjugates. In the mixture, individual mono-polymer-IFN conjugates are defined as positional isomers, depending upon which amino acid residue is covalently attached to the polymer. Within this mixture is an isomer which is an alpha interferon covalently conjugated to a polymer at a histidine residue on the alpha interferon. The compositions are distinguishable from prior art products in part due to the fact that at least about 15%, and preferably at least about 30%, of the interferon conjugates included as part of the composition have a polymer covalently attached to a histidine of the alpha interferon. Preferably, however, the conjugates or positional isomers contain about one polymer strand per alpha interferon, regardless of where the polymer is attached.

Still further aspects of the invention include methods of preparing alpha-interferon conjugates and compositions prepared by the methods. The IFN-polymer conjugates are prepared by reacting a solution containing alpha interferon with a sufficient amount of an oxycarbonyl-N-dicarboximide-activated polymer such as a succinimidyl carbonate activated PEG under conditions which are sufficient to effect covalent attachment of the polymer to the interferon, at least in part, to a His residue such as the His34 of alpha interferon. Part of these conditions include conducting the conjugation reaction within a pH range which is sufficient to facilitate covalent attachment of at least a portion of the polymer strands to histidine residue amino groups of the interferon molecules.

Suitable alpha-interferons include recombinant and non-recombinant alpha-interferons isolated from mammals. The polymer portion of the conjugate is preferably a polyalkylene oxide (PAO), such as a monomethoxy polyethylene glycol (mPEG). In alternative embodiments, other substantially non-antigenic polymers can also be used. The polymers preferably have a molecular weight of from about 200 to about 35,000.

The conditions for effecting conjugation include conducting the attachment reaction with from about an equi-molar to about a relatively small molar excess of the activated polymer with respect to the alpha-interferon. The conditions further include conducting the reaction at a pH of less than about 7 and preferably at a pH of from about 4.5 to about 6.8.

The invention also includes methods of treating alpha-interferon susceptible conditions in mammals. In this aspect, the treatment includes administering an effective amount of the composition containing the IFN conjugates described herein to mammals requiring such therapy.

As a result of the present invention, it has been unexpectedly found that additional improvements in interferon-

polymer conjugate compositions are possible. For example, by modifying the conjugation conditions, it is now possible to obtain compositions containing relatively high activity mono-polymer IFN conjugates in which a portion of the alpha interferon is attached at unique locations to polymers. In addition, it has been found that conducting the conjugation reaction with succinimidyl carbonate and some related oxycarbonyl-N-dicarboximide-type activated polymers, such as SC-PEG, at pH levels which are more acidic than that typically used for conjugation, will cause the polymer to attach not only at the expected lysine sites on the IFN molecule, but also selectively on histidine sites such as the preferred His34 amino acid on alpha interferons.

For a better understanding of the present invention, reference is made to the following description and drawings.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. **1** is a series of chromatograms referred to in Example 11.

FIG. **2** is a series of chromatograms referred to in Example 13.

DETAILED DESCRIPTION OF THE INVENTION

1. Interferons

The interferon (IFN) portion of the polymer conjugate can be prepared or obtained from a variety of sources including recombinant techniques such as those using synthetic genes expressed in *E coli.* See also Pestka, "Interferon α" in *Human Cytokines,* Blackwell Scientific Publications 1–16 (1992), the disclosure of which is incorporated herein by reference. In addition, the IFN can also be a mammalian source extract such as human, ruminant or bovine αIFN. One particularly preferred IFN is IFNα-2b, a recombinantly-made product of the Schering Corp., Kenilworth, N.J.

The term "interferon" or "EFN" as used herein means the family of highly homologous species-specific proteins that inhibit viral replication and cellular proliferation and modulate immune response. Human interferons are grouped into three classes based on their cellular origin and antigenicity: α-interferon (leukocytes), β-interferon (fibroblasts) and γ-interferon (B cells). Recombinant forms of each group have been developed and are commercially available. Subtypes in each group are based on antigenic/structural characteristics. At least 24 interferon alphas (grouped into subtypes A through H) having distinct amino acid sequences have been identified by isolating and sequencing DNA encoding these peptides. See also Viscomi, 1996 *Biotherapy* 10:59–86, the contents of which are incorporated herein by reference. The terms "α-interferon", "alpha interferon", "interferon alpha" and "human leukocyte interferon" are used interchangeably in this application to describe members of this group. Both naturally occurring and recombinant α-interferons, including consensus interferon such as that described in U.S. Pat. No. 4,897,471, the contents of which are incorporated herein by reference, may be used in the practice of the invention.

The purification of interferon alpha from human leukocytes isolated from the buffy coat fraction of whole blood is described in U.S. Pat. No. 4,503,035. Human leukocyte interferon prepared in this manner contains a mixture of human leukocyte interferons having different amino acid sequences. Purified natural human α-interferons and mixtures thereof which may be used in the practice of the invention include but are not limited to Sumiferon® interferon alpha-n1 available from Sumitomo, Japan, Welfferong interferon alpha-n1 (Ins) available from Glaxo-Wellcome Ltd., London, Great Britain, and Alferon® interferon alpha-n3 available from the Purdue Frederick Co., Norwalk, Conn.

The advent of recombinant DNA technology applied to interferon production has permitted several human interferons to be successfully synthesized, thereby enabling the large-scale fermentation, production, isolation, and purification of various interferons to homogeneity. Recombinantly produced interferon retains its in vitro and in vivo antiviral and immunomodulatory activities. It is also understood that the recombinant techniques could also include a glycosylation site for addition of a carbohydrate moiety on the recombinantly-derived polypeptide.

The construction of recombinant DNA plasmids containing sequences encoding at least part of human leukocyte interferon and the expression in *E. coli* of a polypeptide having immunological or biological activity of human leukocyte interferon is disclosed in U.S. Pat. No. 4,530,901 and European Patent No. EP 0 032 134. The construction of hybrid α-interferon genes containing combinations of different subtype sequences (e.g., A and D, A and B, A and F) is disclosed in U.S. Pat. Nos. 4,414,150, 4,456,748 and 4,678,751. Typical suitable recombinant α-interferons which may be used in the practice of the invention include but are not limited to interferon alpha-2b such as Intron® A available from Schering Corporation, Kenilworth, N.J., interferon alpha-2a such as Roferon® A available from Hoffiann-La Roche, Nutley, N.J., and Infergen® available form Amgen, Thousand Oaks, Calif.

Alternate embodiments, where the foreign αIFN is not completely autologous, may be also used if desired. A key, however, is that the non-autologous αIFN has sufficient bioactivity or αIFN effect such as antiviral activity in the target mammal. Other substances including αIFN fractions or predecessor polypeptides can also be included in the conjugates of the present invention. As used herein, "α-IFN effect in mammals" means in vivo activity corresponding to that observed with αIFNs. These substances are prepared by using techniques known to those of ordinary skill in the art such as tissue culture, extraction from animal sources or by recombinant DNA methodologies. Transgenic sources of αIFN and related moieties are also contemplated. Such materials are obtained from transgenic animals, i.e. mice, pigs, cows, etc. where the αIFN protein is expressed in milk, blood, or other tissues. The method by which the αIFN is prepared for the conjugates of the present invention is not limited to those described herein. For purposes of the present invention, the αIFN's are preferred because of their biochemical and serological properties. In particular, αIFN has documented antiviral properties and diffuses more effectively into the bloodstream than other interferons.

2. Non-Antigenic Polymers

To conjugate the IFN to polymers such as poly(alkylene oxides), one of the polymer hydroxyl end-groups is converted into a reactive functional group which allows conjugation. This process is frequently referred to as "activation" and the product is called an "activated" polymer or activated poly(alkylene oxide). Other substantially non-antigenic polymers are similarly "activated" or functionalized.

The activated polymers are reacted with αIFN so that attachment occurs at ϵ-amino groups of lysines, the N-terminal cysteine amino group and, as described below, at amino groups on histidines. Free carboxylic acid groups, suitably activated carbonyl groups, oxidized carbohydrate moieties and mercapto groups if available on the IFN can also be used as supplemental attachment sites, if desired.

In a preferred aspect of the invention, urethane (carbamate) linkages are formed between one of the αIFN amino acid amino groups (i.e. lysine, histidine, N-terminal), and the activated polymer. Preferably, the urethane linkage is formed using a terminal oxycarbonyl-oxy-N-dicarboximide group such as a succinimidyl carbonate group. Alternative activating groups include N-succinimide, N-phthalimide, N-glutarimide, N-tetrahydrophthalimide and N-norborene-2,3-dicarboxide. These urethane-forming groups are described in commonly owned U.S. Pat. No. 5,122,614, the disclosure of which is hereby incorporated by reference. This patent also discloses the formation of N-succinimide carbonate derivatives of polyalkylene oxides including polyethylene glycols which are also capable of forming urethane linkages with lysine amino group targets.

Among the substantially non-antigenic polymers, mono-activated, alkoxy-terminated polyalkylene oxides (PAO's), such as monomethoxy-terminated polyethylene glycols (mPEG's) are preferred; bis-activated polyethylene oxides (glycols) are also contemplated for purposes of cross-linking αIFN's or providing a means for attaching other moieties such as targeting agents for localizing the polymer-αIFN conjugate in a particular area such as, for example, the liver.

Suitable polymers will vary substantially by weight. Polymers having molecular number average weights ranging from about 200 to about 35,000 are usually selected for the purposes of the present invention. Molecular weights of from about 1,000 to about 15,000 are preferred and 2,000 to about 12,500 are particularly preferred.

The polymeric substances included are also preferably water-soluble at room temperature. A non-limiting list of such polymers include polyalkylene oxide homopolymers such as polyethylene glycol (PEG) or polypropylene glycols, polyoxyethylenated polyols, copolymers thereof and block copolymers thereof, provided that the water solubility of the block copolymers is maintained. In addition to mPEG, C_{1-4} alkyl-terminated polymers are also useful.

As an alternative to PAO-based polymers, effectively non-antigenic materials such as dextran, polyvinyl pyrrolidones, polyacrylamides such as HPMA's-hydroxypropylmethacrylamides, polyvinyl alcohols, carbohydrate-based polymers, copolymers of the foregoing, and the like can be used. Those of ordinary skill in the art will realize that the foregoing list is merely illustrative and that all polymer materials having the qualities described herein are contemplated. For purposes of the present invention, "substantially or effectively non-antigenic" means all materials understood in the art as being nontoxic and not eliciting an appreciable immunogenic response in mammals.

3. Reaction Conditions

Conjugation reactions, sometimes referred to as PEGylation reactions, are often carried out in solution without regard to where the polymer will attach to the protein. Such techniques are also usually carried out at slightly alkaline i.e. pH 7+ to about 9 for conjugating αIFNs. A key to the present invention, however, is that the retained IFN bioactivity can be maximized if the polymer is attached to a histidine, preferably His34 on IFNα 2b. It will be appreciated by the artisan that although various species of the αIFN may or may not have a histidine at amino acid 34, the interferon conjugates will nonetheless preferably include at least some positional isomers containing a polymer attached at an available histidine.

The processes of the present invention therefore includes reacting a solution containing an alpha interferon with an amount of an oxycarbonyl-oxy-N-dicarboximide-activated polymer such as succinimidyl carbonate-activated mPEG at a pH which is sufficient to facilitate covalent attachment of at least a portion of the polymer strands to a histidine, such as the His34 of IFNα2b, of the individual interferon molecules. In particular, the pH will preferably be slightly acidic, i.e. less than about 7.0; more preferably, less than about 6.8 and most preferably in the range of from about 4.5 to about 6.8.

The reaction conditions for effecting conjugation further include conducting the attachment reaction with from about equi-molar to about a relatively small molar excess of the activated polymer with respect to the alpha-interferon. In this regard, the process can be carried out with about 1–8-fold molar excesses; preferably about 1.5–7-fold molar excesses and most preferably about 1.75–5-fold molar excesses. The conjugation reaction can be carried out at about room temperature, 20–25° C. It is also preferred that the coupling reaction be allowed to proceed for rather short periods of time, i.e. 1–2 hours, before quenching. In practice, the reaction conditions provide a mixture of polymer-IFN positional isomers. Preferably, each isomer contains a single polymer strand attached to the interferon via an amino acid residue. In alternative embodiments, there can be more than one strand of polymer attached as a result of the process. Solutions containing these conjugates are also useful as is or can be further processed to separate the conjugates on the basis of molecular weight.

Characterization of the preferred one polymer strand-IFN conjugates (isomers) via cation exchange chromatography into separated peaks revealed that the polymer can be attached at up to about eight different sites on the IFNα2b molecule. These sites, representing individual positional isomers, are Cys1, Lys31, His34, Lys49, Lys83, Lys121, Lys131, Lys134. In some preferred embodiments, the reaction pools containing mono-polymer-IFN conjugates can contain relatively high proportions of the His34 positional isomer, i.e. about 30–60%, the Cys1 positional isomer, about 7–20%, and the Lys121 positional isomer, about 7–15%, with the rest of the positional isomers comprising the remainder. It will be understood that alternative IFN's will provide alternative distributions of positional isomers, depending upon the amino acid sequence of the starting material.

Due to the nature of the solution-based conjugation reactions, the compositions are a heterogeneous mixture of species which contain the polymer strand(s) attached at different sites on the interferon molecule. In any solution containing the conjugates, it is likely that a mixture of at least about 3, preferably about 6 and more preferably about 8 positional isomers will be present. For example, when IFNα2b is used, the solution will contain conjugate isomers with the polymer attached at one or more of Cys1, Lys31, His34, Lys49, Lys83, Lys121, Lys131, and Lys134 of the interferon. In the case of IFNα2b and the preferred forms of activated polymers described herein, the 3 most prominent sites of attachment are His34 (55%), Cys1 (15%) and Lys121(15%).

A preferred composition of the invention is a mixture of the IFN-polymer isomers which are composed of at least about 15% His-polymer substituted-IFN. That is, at least about 15% of the conjugates include covalent attachment of the alpha interferon to the substantially non-antigenic polymer at a His. In more preferred aspects, at least about 30%, and in most preferred aspects of the invention, at least about 40% of the conjugates include the His34 covalent polymer attachment. When IFNα2b or related IFN's are used, the histidine attachment site is preferably His34.

4 Effect of Reaction pH upon PEG-IFN Positional Isomers Distribution

The process of the present invention takes advantage of the discovery that the site of polymer attachment on alpha interferon is influenced to a large extent by the pH of the reaction system. As the pH of the reaction solution is varied, the reactivity towards specific forms of activated polymers of the various functional groups such as alpha-amines, imidazoles and epsilon amines will vary. Typically, polymer conjugation reactions are carried out at basic pHs in order to maximize attachment at lysine epsilon amino groups. For example, Zalipsky et al. *Biotech. & App. Biochem,* Vol 15, p. 100–114; (1992) evaluated the SC-PEG reagent for PEGylation and reported that the optimal reactivity was at about pH 9.3. The method of the present invention, however, includes conducting the reaction at lower pH's in order to allow a portion of the activated polymer strands to attach to histidine amino groups and de-emphasize, but not eliminate, lysine sites for attachment.

Furthermore, it has also been found that the biological activity of the various polymer conjugate positional isomers unexpectedly differs, even when each of the positional isomers has the same degree of polymer substitution.

The method described herein affords novel attachment of polymers such as PEG to a specific histidine residue in IFN molecules. In preferred embodiments, the conjugation reaction results in a substantial amount, i.e. at least about 30% of the conjugates being linked at IFN histidine sites such as the His34 on IFNα2b.

It has also been unexpectedly determined that the relative distribution of the positional isomers is largely dependent upon the pH at which the conjugation reaction is carried out. Shifting the pH from basic to slightly acidic pH (5.5–6.5) favors the formation of conjugates linked at His34 on IFNα2b, and to a lesser extent, the N-terminus (Cys1). Using pH(8–10) during the conjugation reaction, on the other hand, favors the formation of lysine-related attachment sites, confirmed via cation exchange chromatography. In those situations where IFNα2b is not included, the His34 site, of course, may not always be present. The reaction conditions nonetheless allow covalent attachment of an activated polymer to a His. Thus, Applicants have demonstrated that the pH of the reaction system influences the placement of some types of activated polymers on a protein surface, especially with regard to different amino acid residues (i.e. lysine vs. N-terminal amine vs. histidine).

5. Fractionation of Conjugates

Although the inventive process produces a substantial amount of conjugates having a single polymer strand, conjugates having varying degrees of polyalkylene oxide substitution are also generated. Residual unconjugated PAO's and αIFN can also be present. This mixture is typically in a reaction buffer containing one or more of phosphate, chloride and bicarbonate anions. The PAO, αIFN and conjugate mixture is preferably fractionated in a buffer solution containing from about 1–10 mg/ml PAO-αIFN conjugates. Suitable fractionating solutions have a pH of from about 7.0 to about 9.0 and preferably from about 7.5 to about 8.5. The solutions preferably contain one or more buffer salts selected from KCl, NaCl, K_2HPO_4, KH2PO4, Na_2HPO_4, NaH_2PO_4, $NaHCO_3$, $NaBO_4$, $(NH_4)_2CO_3$ and glycine NaOH. Sodium phosphate buffers are preferred.

Depending upon the reaction buffer, the αIFN-polymer conjugate containing solution may first have to undergo buffer exchange/ultrafiltration. For example, the αIFN conjugate solutions can be ultra filtered across a low molecular weight cut-off (10,000 to 30,000 Dalton) membrane which will also remove most surfactants, if present, as well.

The fractionation of the conjugates into desired species is preferably carried out using an anion exchange medium. Such media are capable of selectively binding those αIFN-polymer conjugates having 1–4 polymer strands, excess polymer and unmodified αIFN. This fractionation occurs since the αIFN molecules of various degrees of substitution will have isoelectric points which vary in a somewhat predictable fashion. For example, the isoelectric point of αIFN is determined by the number of available amino groups available on the surface of the protein. These amino groups also serve as the point of attachment of polyalkylene oxide conjugates. Therefore, as the degree of substitution of polyalkylene oxide increases, the isoelectric point decreases, and the ability of the conjugate to bind to an anion exchange resin weakens.

The use of strongly polar anion exchange resins is especially preferred for the method of the present invention. For this reason, quaternary amine coated anion exchange resins are utilized. The quaternary amine resin may be coated onto either a polymeric or silica matrix; however, polymeric matrices are preferred. A number of tetramethylamine, or quaternary methylamine, anion exchange resins are commercially available, coated onto the support matrices. Included among the commercially available quaternary anion exchange resins suitable for use with the present invention are Q-HD available from Bio-Sepra; QA TRISACRYL® and QMA-SPHEROSIL®, quaternary amine resins coated onto a polymer matrix, manufactured by IBF of Garenne, France, for Sepracor, Inc. of Marlborough, Mass.; TMAE650M®, a tetramethylamino ethyl resin coated onto a polymer matrix, manufactured by EM-Separators of Gibbstown, N.J.; QAE550C®, and SUPERQC®, each a quaternary amine resin coated onto a polymer matrix and manufactured by TosoHaas of Montgomeryville, Pa. QMA Accell, manufactured by Millipore of Millford, Mass. and PEI resins manufactured by J T Baker of Phillipsburg, N.J., may also be used.

The anion exchange resin is packed in the column and equilibrated by conventional means. A buffer having the same pH and osmolality as the conjugated αIFN solution is used. The conjugate-containing solution is then adsorbed onto the column. At the completion of the loading, a gradient flow of an elution buffer with increasing salt concentrations is applied to the column to elute the desired fractions of polyalkylene oxide-conjugated αIFN. The fractions are of essentially uniform molecular weight and degree of substitution.

Preferred IFN conjugate fractions have 1–4 polymer strands per αIFN molecule. More preferably, the fraction contains about 1–2 and, most preferably, about 1 polymer strand per αIFN molecule. The elution buffer preferably contains one or more salts selected from KCl, NaCl, K_2HPO_4, KH_2PO_4, NA_2HPO_4, NaH_2PO_4, $NaHCO_3$, $NaBO_4$ and $(NH4)C_3$. These fractions are substantially free of other conjugates. Any unconjugated species can then be backwashed from the column by conventional techniques.

Techniques utilizing multiple isocratic steps of increasing concentration can also be used. Multiple isocratic elution steps of increasing concentration will result in the sequential elution of αIFN-polymer conjugates. The degree of polymer conjugation within each fraction will be substantially uniform. However, the degree of polymer conjugation for each fraction will decrease with elution time. Ion exchange purification of the conjugates can also be carried out with, for

example, a Q-HD Column from Sepracor, Inc. along with a dilute sodium phosphate solution (10 mM $NaPO_4$ ion). The sample is washed with 10 mM $NaPO_4$ to remove any unreacted PAO and thereafter a step gradient elution with NaCl is used. Elution with 10 mM NaCl recovers fractions containing conjugates with greater than 3 polymer strands PAO per IFN; elution with 50 mM NaCl recovers conjugates containing 1–2 strands; elution with 150 mM NaCl recovers unmodified IFN.

The temperature range for elution is between about 4° C. and about 25° C. Preferably, elution is carried out at a temperature of from about 6° C. to about 22° C. The elution of the PAO-αIFN fraction is detected by UV absorbance at 254 nm. Fraction collection may be achieved through simple time elution profiles. The preferred fractions can also be pooled in the elution buffer.

6. Surfactants

In another preferred aspect, the reaction conditions include the presence of a surfactant. The surfactants used in the processes of the present invention are ionic-type agents. One particularly preferred agent is sodium dodecyl sulfate, (SDS). Other ionic surfactants such as lithium dodecyl sulfate, quaternary ammonium compounds, taurocholic acid, caprylic acid, decane sulfonic acid, etc. can also be used. Non-ionic surfactants can also be used. For example, materials such as polyoxyethylene sorbitans (TWEEN®'s), polyoxyethylene ethers (Tritons) can be used. See also Neugebauer, *A Guide to the Properties and Uses of Detergents in Biology and Biochemistry* (1992) Calbiochem Corp. The only limitations on the type of surfactant used in the processes of the invention are that they do not cause substantial denaturation of the IFN and do not completely inhibit polymer conjugation. The surfactants are present in the reaction mixtures in amounts from about 0.01–0.5%; preferably from 0.05–0.5%; and most preferably from about 0.075–0.25%. Mixtures of the surfactants are also contemplated.

7. Pharmacokinetic Parameters

As pointed out above, compositions of the present invention contain a heterogeneous mixture of polymer-IFN species in which the polymer strand(s) is/are attached at different sites on the interferon molecule. In spite of the heterogeneous nature of the conjugates, the compositions have a predictable in vivo pharmacokinetic profile which maximizes the therapeutic effect of the interferon.

Compositions of the present invention containing IFNα preferably include at least about 15% polymer-His conjugates, more preferably at least about 30% and most preferably at least about 40% polymer-His conjugates. While Applicants are not bound by theory, it is believed that the linkage for the His-positional isomers included in the compositions of the invention is relatively labile vis a vis that of the Lys-positional isomers. As a result, at physiologic pH, the compositions demonstrate a relatively smooth onset on activity after administration as well as a prolonged duration of effect. This profile allows the artisan to administer the composition in less frequent doses than with unmodified IFN's.

8. Methods of Treatment

Another aspect of the present invention provides methods of treatment for various medical conditions in mammals, preferably humans. The methods include administering an effective amount of an αIFN-polymer conjugate containing composition which has been prepared as described herein to a mammal in need of such treatment. The conjugates are useful for, among other things, treating interferon-susceptible conditions or conditions which would respond positively or favorably as these terms are known in the medical arts to interferon-based therapy.

Conditions that can be treated in accordance with the present invention are generally those that are susceptible to treatment with interferon alpha For example, susceptible conditions include conditions which would respond positively or favorably as these terms are known in the medical arts to interferon alpha-based therapy. For purposes of the invention, conditions that can be treated with interferon alpha therapy include those conditions in which treatment with an interferon alpha shows some efficacy, but which may not be treatable with interferon alpha because the negative side effects outweigh the benefits of the treatment. For example, side effects accompanying alpha therapy have virtually ruled out treatment of Epstein Barr virus using interferon alpha. Practice of the invention results in substantially reduced or eliminated side effects as compared to conventional interferon alpha treatment.

Exemplary conditions which can be treated with interferon include but are not limited to cell proliferation disorders, in particular cancer (e.g., hairy cell leukemia, Kaposi's sarcoma, chronic myelogenous leukemia, multiple myeloma, basal cell carcinoma and malignant melanoma, ovarian cancer, cutaneous T cell lymphoma), and viral infections. Without limitation, treatment with interferon may be used to treat conditions which would benefit from inhibiting the replication of interferon-sensitive viruses. Viral infections which may be treated in accordance with the invention include hepatitis A, hepatitis B, hepatitis C, other non-A/non-B hepatitis, herpes virus, Epstein-Barr virus (EBV), cytomegalovirus (CMV), herpes simplex, human herpes virus type 6 (HHV-6)), papilloma, poxvirus, picomavirus, adenovirus, rhinovirus, human T lymphotropic virus-type 1 and 2 (HTLV-1/-2), human rotavirus, rabies, retroviruses including human immunodeficiency virus (HIV), encephalitis and respiratory viral infections. The method of the invention can also be used to modify various immune responses.

Variants of interferon alpha are currently approved in the United States and other countries for the treatment of hairy cell leukemia, venereal warts, Kaposi's Sarcoma, and chronic non-A/non-B hepatitis: interferon alpha-2b, marketed under the trade name INTRON® A (Schering Corporation, Kenilworth N.J.), and interferon alpha-2a, marketed under the trade name Roferon® A (Hoffmann-La Roche, Nutley, N.J.), and consensus interferon marketed under the trade name Infergen™ (Amgen, Thousand Oaks, Calif.). Since interferon alpha-2b, among all interferons, has the broadest approval throughout the world for treating chronic hepatitis C infection, it is most preferred for use in the treatment of chronic hepatitis C in accordance with practice of the invention.

Administration of the described dosages may be every other day, but is preferably once or twice a week. Doses are usually administered over at least a 24 week period by injection.

Administration of the dose can be intravenous, subcutaneous, intramuscular, or any other acceptable systemic method. Based on the judgment of the attending clinician, the amount of drug administered and the treatment regimen used will, of course, be dependent on the age, sex and medical history of the patient being treated, the neutrophil count (e.g. the severity of the neutropenia), the severity of the specific disease condition and the tolerance of the patient to the treatment as evidenced by local toxicity and by

systemic side-effects. Dosage amount and frequency may be determined during initial screenings of neutrophil count.

Conventional pharmaceutical formulations can be also prepared using the conjugate-containing compositions of the present invention. The formulations comprise a therapeutically effective amount of the interferon-polymer conjugate composition together with pharmaceutically acceptable carriers. For example, adjuvants, diluents, preservatives and/or solubilizers, if needed, may be used in the practice of the invention. Pharmaceutical compositions of interferon including those of the present invention may include diluents of various buffers (e.g., Tris-HCl, acetate, phosphate) having a range of pH and ionic strength, carriers (e.g., human serum albumin), solubilizers (e.g., polyoxyethylene sorbitan or TWEEN®, polysorbate), and preservatives (e.g., thimerosol, benzyl alcohol). See, for example, U.S. Pat. No. 4,496,537.

The amount of the α-IFN polymer conjugate administered to treat the conditions described above is based on the IFN activity of the polymeric conjugate. It is an amount that is sufficient to significantly affect a positive clinical response. Although the clinical dose will cause some level of side effects in some patients, the maximal dose for mammals including humans is the highest dose that does not cause unmanageable clinically-important side effects. For purposes of the present invention, such clinically important side effects are those which would require cessation of therapy due to severe flu-like symptoms, central nervous system depression, severe gastrointestinal disorders, alopecia, severe pruritus or rash. Substantial white and/or red blood cell and/or liver enzyme abnormalities or anemia-like conditions are also dose limiting.

Naturally, the dosages of the various αIFN compositions will vary somewhat depending upon the αIFN moiety and polymer selected. In general, however, the conjugate is administered in amounts ranging from about 100,000 to about several million IU/m^2 per day, based on the mammal's condition. The range set forth above is illustrative and those skilled in the art will determine the optimal dosing of the conjugate selected based on clinical experience and the treatment indication.

The pharmaceutical compositions may be in the form of a solution, suspension, tablet, capsule, lyophilized powder or the like, prepared according to methods well known in the art. It is also contemplated that administration of such compositions will be chiefly by the parenteral route although oral or inhalation routes may also be used depending upon the needs of the artisan.

EXAMPLES

The following examples serve to provide further appreciation of the invention but are not meant in any way to restrict the effective scope of the invention.

Example 1

Preparation of rαIFN-$PEG_{5,000}$ in presence of SDS (0.1%)

In this example, recombinant αIFN-2b, (rαIFN), a product of the Schering-Plough Corporation, Kenilworth, N.J. was conjugated with activated polyethylene glycol-N-succinimide carbonate (SC-PEG) as described in U.S. Pat. No. 5,122,614. The polymer had a molecular weight of about 5,000.

36 mg of the rαIFN was dialyzed into 0.1 molar sodium phosphate pH 7.5 using a Centricon-10 (a product of the Amicon Corporation of Beverly, Mass.). The final concentration of rαIFN was about 3 mg/ml. 0.1 ml of 10% SDS was added to the rαIFN and was allowed to incubate at room temperature for 10 minutes. Thereafter, 42 mg of SC-$PEG_{5,000}$ was added to the protein-SDS solution and stirred at room temperature for two hours and then quenched with glycine. Next, the reaction mixture was dialyzed into 10 mM sodium phosphate pH 8 to fractionate the PEGylated IFN using a Centricon-30.

Example 2

Preparation of rαIFN-$PEG_{12,000}$ in presence of SDS (0.1%)

In this Example, the steps of Example 1 were repeated except that the polyethylene glycol had a molecular weight of about 12,000. Reaction steps were exactly the same to provide the $PEG_{12,000}$ conjugate.

Example 3

Fractionation of $2PEG_{5,000}$rαIFN

In this Example the conjugates prepared in accordance with Example 1 were fractionated to obtain the desired 2-$PEG_{5,000}$ fraction. The PEG-αIFN in sodium phosphate buffer was loaded onto a QHD anion exchange column. The 2-PEG fraction was eluted with a gradient from 0 to 400 mM sodium chloride in 10 mM phosphate pH 8. The 2-PEG fraction was verified using size exclusion chromatography and SDS-PAGE.

Example 4

Fractionation of $2PEG_{12,000}$rαIFN

The polymer conjugates of Example 2 were fractionated in the manner described in Example 3 and verified in the same manner.

Examples 5–8

In these examples, additional preparations of $PEG_{12,000}$-rαIFN were prepared as described previously except that no surfactant was used. Following the conjugation reactions, the samples were tested for retained activity and PEG number. The results are provided below in the table.

TABLE 1

IFN-$PEG_{12,000}$ PREPARATION	ACTIVITY (CPE) % OF CONTROL	PEG #
Example 6	26	1.2
Example 7	26	1.3
Example 8	24	1.0

Example 9

Comparative Data

In this example, the product of Example 3, (SDS-2-$PEG_{5,000}$rαIFN), 2-$PEG_{5,000}$rαIFN made in the absence of a surfactant and unconjugated rαIFN were tested. Activity was determined using a CPE assay with EMC virus challenging A549 human lung carcinoma cells. Circulating life was determined using an average value obtained from the blood of 3 rats in a group receiving 1 million units, with time points taken over 7 days.

TABLE 2

	ACTIVITY (%)	VIRAL PROTECTION ASSAY IC_{50} (pg/ml)	CIRCULATING HALF LIFE α PHASE (HRS.)
A. IFN-SDS 2-$PEG_{5,000}$	69	2.2	5.8
B. IFN-$PEG_{5,000}$	30	4.0	6.8
C. IFN	100	1.5	0.17

This data clearly shows the advantages of the inventive process. Retained activity is over twice as great as that obtained using standard techniques.

Example 10

In this example, various pharmacokinetic data was generated using 2PEG-rαIFN conjugates prepared according to the methods described above. These samples were compared to unmodified IFN according to the protocol set out in Table 4. Sample B was prepared with SDS.

TABLE 3

	Retained Activity	
SAMPLE	PEG MOLECULAR WEIGHT	CPE ACTIVITY (% CONTROL)
A	5,000	35
B	5,000	69
C	12,000	26
D	12,000	26

For example:

TABLE 4

Pharmacokinetic Protocol	
ANIMALS:	Sprague Dawley (3 rates/time point)
DOSE:	10×10^6 UN IFN/rat
ROUTE:	Subcutaneous (S.C.)
DRUG:	2-PEG-IFNα's 5,000 and 12,000 mol. wt. PEG
TIME POINTS:	0 min., 5 min., 15 min., 30 min., 1 hr., 2 hr., 4 hr., 8 hr., 24 hr., 48 hr., 5 days, and 7 days following drug administration.
ASSAY:	CPE Assay using serum samples in an EMC virus and A549 human lung carcinoma.
AUC =	Area Under Curve, C_{max}, $T_{½}\alpha$, $T_{½}\beta$- all have their generally ascribed meanings known to those of ordinary skill.

Tables 5 and 6

Summary of Pharmacokinetics Data for PEG-Interferons

TABLE 5

SAMPLE	IC_{50} (pg/ml)	% ACTIVITY	AUC	Cmax (IU/ml)
NATIVE IFNα	1.52 pg/ml (N = 6)	100%	145,720	60,000
A	4.0 pg/ml (N = 3)	35%	348,920	24,433
B	2.2 ± 0.5 pg/ml (N = 3)	69%	351,037	—
C	5.8 ± 2.2 pg/ml (N = 3)	26%	1,574,682	62,750

TABLE 6

SAMPLE	T_{max} (hr)	$T_{½}\alpha$ PHASE (hr)	$T_{½}\beta$ PHASE (HR)
NATIVE IFNα	1	0.17	—
A	4	6.8	48
B	2–3	5.8	—
C	8	12.1	33

The foregoing data provide the following conclusions:

2-PEG-rαIFN conjugates prepared with both 5,000 and 12,000 molecular weight have distinct advantages over unmodified interferon in mammals. In the case of subcutaneously administered compositions, T_{max} is substantially increased by the conjugation of the protein with about 2 PEG's. For chronic conditions, longer T_{max}'s are desirable and allow clinicians to space out recurring administrations due to the lengthening of the duration of effect. Even more unexpected, however, was the fact that 2-$PEG_{12,000}$ conjugates are able to unexpectedly increase AUC by over 10-fold. This dramatic increase in area under the curve was not proportional to the additional polymer weight. Clearly, therapeutic advantages are realized by this unexpected increase.

Example 11

Effect of pH on PEGylation

In order to probe this effect, the polymer conjugation (PEGylation) reaction of Examples 5–8 was repeated using $mPEG_{12,000}$ (no surfactant) at four different pHs, 5.4, 6.5, 8.0 and 10.0. The ratio of 2.6 grams of SC-$PEG_{12,000}$ to 1 gram of IFN (molar ratio 3.9:1) was used for the reactions at pH 5.4, 6.5 and 8.0 while the ratio of 2.1 grams of SC-$PEG_{12,000}$ to 1 gram of IFN (molar ratio 3.2:1) was used at pH 10. At the end of the reaction, glycine was added to quench any residual PEGylation reagent. The product from each reaction was then purified using a Q-hyper D resin at pH 8 with salt elution to remove unreacted ingredients.

The purified conjugates obtained at the different pHs were evaluated for their biological activity, hydroxylamine sensitivity and distribution of positional isomers. Biological activity was determined by specific activity (MTT-CPE assay).

Hydroxylamine sensitivity was undertaken to determine what percentage of the conjugates were PEGylated at histidine sites, including the IFN-His34. Hydroxylamine is a known reagent that we have found to selectively cleave PEG from IFN histidines. An aliquot of each of the samples (50 μl) was diluted with 0.45 ml of 10 mM sodium phosphate pH 7.0 An aliquot of this protein solution (150 μl) was treated with 150 μl of 0.5 M hydroxylamine and incubated at room temperature for 60 minutes. Thereafter, a volume of 75 μl was loaded on a Mini-S column (Pharmacia Biotech) for cation exchange chromatography. Mobile phase A included 10 mM sodium acetate pH 5.3 buffer and 25% 2-propanol. Mobile phase B contained 500 mM sodium chloride dissolved in mobile phase A. The flow rate was set at 0.5

ml/min and the eluted protein was detected at 214 nm. The individual PEG-IFN solutions were diluted with 10 mM sodium acetate pH 5.3, containing 2-propanol (5%) to 1 mg/ml protein concentration. Injection volumes ranged from 10 to 30 μl, depending upon the protein concentration. A linear gradient was used. The results are set forth in the Table 7 below and in FIG. 1.

FIG. 1 shows the overlay of the chromatograms obtained from the Mono-S cation exchange chromatography column of the different pH reaction products. The site of polymer conjugation for each positional isomer was determined by digestion of individual peaks from cation exchange chromatography using proteolytic enzymes (trypsin, V8-protease, chymotrypsin or subtilisin), isolation of PEGylated fragments, and analysis by N-terminal sequencing and mass spectroscopy.

As seen in the figure, the distribution of the positional isomers changes significantly as the pH of the reaction changes. The higher the pH, the less His34-linked PEG-IFN and, less dramatically, Cys1-linked PEG-IFN products are produced.

Table 7 summarizes the specific bioactivity as determined using the MT-CPE bioassay for IFN and the amount of IFN released upon treatment with 0.5M hydroxylamine for 2 hours at 25° C. for the different conjugate products. These findings confirm that the differences seen in FIG. 1 can also be related to different biological characteristics of the products. When the conjugation is conducted at a higher pH (i.e. 8 or 10) the products formed are less bioactive and more resistant to hydroxylamine, which therefore means that at higher pH's, less polymer is on His34.

TABLE 7

Bioactivities and Hydroxylamine Sensitivities of PEG-IFNs Generated at Different pHs

Reaction pH	Specific Activity (CPE assay) MIU/mg	% of Conjugate Converted to IFN by Hydroxylamine
5.4	61.8	56%
6.5	74.5	47%
8.0	33.3	8%
10.0	27.8	<1%

The above results indicate that pH is a key variable of the conjugation reaction and that the relative distribution of the positional isomers varies dramatically with pH. Unexpectedly, the bioactivity of the resultant PEG-IFN mixture of positional isomers is it also affected.

Example 12

Comparison of Urethane Linkage Forming Activated Polymers

In this example, effect of pH on reaction conditions was compared using a different type of urethane linker to see if the activating group had any role in determining the site of polymer attachment and bioactivity. In particular, the Methoxypoly(ethylene glycol)-succinimidyl carbonate MW 12,000 (SC-$PEG_{12,000}$) used in the earlier examples was compared with methoxypoly(ethylene glycol)-2-pyridyl carbonate, MW 12,000 (PC-$PEG_{12,000}$) disclosed in U.S. Pat. No. 5,382,657, as the activated polymer reagents for interferon alpha-2b (IFN). The conjugation reactions were carried out for both reagents, SC-$PEG_{12,000}$ and PC-$PEG_{12,000}$, at pH 6.5 and 10.0. The conditions used to generate the 4 monopegylated IFN samples for analysis were 1) SC-$PEG_{12,000}$ @ pH 6.5; 2) PC-$PEG_{12,000}$ @ pH 6.5; 3) SC-$PEG_{12,000}$ @ pH 10.0; and 4) PC-$PEG_{12,000}$ @ pH 10.0. In each pH 6.5 case, a 3.9 to 1 molar ratio of PEG: IFN was used. In each pH 10.0 case, a 3.2:1 molar ratio of PEG:IFN was used. These conditions were chosen to evaluate the influence of both reaction pH and linker on the composition of the final product.

The conjugated material from each reaction condition was recovered and tested for biological activity (CPE assay) and for distribution of positional isomers using Mini-S chromatography assay.

The PEG-IFN generated by reacting IFN with PC-$PEG_{12,000}$ at pH 6.5 had lower biological activity than that made with SC-$PEG_{12,000}$ in spite of both reagents forming urethane bonds. Thus, it was shown that in spite of the similarity between the linkers, SC-PEG, an oxycarbonyl-oxy-N-dicarboximide-activated polymer, more preferentially attaches to His34. Interestingly, however, the PEG-IFN products generated by carrying out the reaction at pH 10 with both PC-$PEG_{12,000}$ and SC-$PEG_{12,000}$ had similar biological activities. In both cases, however, the activities were lower than that obtained for SC-$PEG_{12,000}$ at pH 6.5.

Mini-S chromatography assays showed that histidine-34-linked PEG-IFN is the major positional isomer present when using SC-$PEG_{12,000}$ at pH 6.5. Lysine-121-linked PEG-IFN is the major positional isomer present when the reaction is carried out at pH 6.5 using PC-$PEG_{12,000}$. At pH 10, Lysine-121-linked PEG-IFN is the major product using either reagent. See Table 8.

Thus, the use of acidic pH and an oxycarbonyl-oxy-N-dicarboximide-activated polymer, i.e. SC-PEG, produce conjugates which are unique products which cannot be reproduced by substituting another urethane bond-forming activated polymer such as PC-$PEG_{12,000}$ in place of SC-$PEG_{12,000}$.

The above materials contained less than 5% total di-PEG and multi-PEG-IFN as indicated by the size-exclusion HPLC assay.

TABLE 8

Summary of MiniS Assay Results

	PEAK NUMBER - (Area Percent)							
Sample	1	2	3	4	5	6	7	8
SC-PEG; pH 6.5	21	63	ND	0.7	11.8	5.6	3.4	13.3
PC-PEG; pH 6.5	ND	4.8	9	9.6	33.8	13	3.8	25.9
SC-PEG; pH 10	ND	ND	14.8	11.2	57.6	9.5	3.1	3.8
PC-PEG; pH 10	ND	ND	9.6	13.8	51.7	13.7	3.5	7.8

ND: not detected

Peak Assignment: Peak 2: His-34 linked PEG-IFN; Peak 4: Lys-31 linked PEG-IFN;
Peak 5: Lys-121 linked PEG-IFN; Peak 6: Lys49 linked PEG-IFN;
Peak 7: Lys-83 linked PEG-IFN; Peak 8: N-terminus (cysteine) linked PEG-IFN

Example 13

Cation Exchange Chromatography Characterization

In this example, analytical separation of several batches of PEG-IFN product produced using the procedure of Example 11 (pH 6.5) was carried out using cation exchange chromatography in order to determine the sites of polymer attach-

ment and identify the individual positional isomers. The cation exchange apparatus was a Mini-S column (Pharmacia Biotech). Mobile phase A included 10 mM sodium acetate pH 5.3 buffer and 25% 2-propanol. Mobile phase B contained 500 mM sodium chloride dissolved in mobile phase A. The flow rate was set at 0.5 ml/min and the eluted protein was detected at 214 nm. The individual PEG-IFN solutions were diluted with 10 mM sodium acetate pH 5.3, containing 2-propanol (5%) to 1 mg/ml protein concentration. Injection volumes ranged from 10 to 30 μl, depending upon the protein concentration. The following linear gradient was used:

Time (min)	A(%)	B(%)
0	100	0
5	93	7
50	83	17
60	0	100
65	0	100
66	100	0
75	100	0

The results are provided in Table 9 below and graphically illustrated in FIG. 2.

TABLE 9

Area Percent Quantification of PEG-IFN Batches by Cation Exchange Chromotography

Batch	Peak 2	Peaks 3/4	Peak 5	Peak 6	Peak 7a	Peak 7b	Peak 8
1	2.6	53.2	5.3	14.2	6.5	3.4	17.2
2	1.5	54.7	3.3	12.6	6.1	3.2	18.6
3	1.6	55.3	2.4	11.9	5.5	3.2	20.1
4	1.7	55.1	2.6	11.6	5.3	3.1	20.5
5	1.7	54.3	2.7	11.8	5.6	3.2	20.7
6	1.7	54.5	2.6	11.8	5.3	2.9	21.1
7	1.9	54.2	2.3	11.6	5.2	3.2	21.5

Main Peak Assignment: Peak 2: Lys-134 linked EPG-IFN; Peak 3/4: His-34 linked PEG-IFN; Peak 6: Lys-121 linked PEG-IFN and Lys-131 linked PEG-IFN; Peak 8: Cys-1 linked PEG-IFN.

These results illustrate that a majority of the conjugates were found in peaks 3 and 4 (His-34 linked PEG-IFN). The results also show that contrary to what was expected, most of the conjugates were formed by attaching the polymer to a histidine rather than one of the lysine amino groups.

Other embodiments of the invention will be apparent to one skilled in the art from a consideration of this specification or practice of the invention disclosed herein. It is intended that the specification and examples be considered as exemplary only, with the true scope and spirit of the invention being indicated by the following claims.

What is claimed is:

1. A pharmaceutical composition, comprising a mixture of alpha interferon polymer conjugate positional isomers, wherein one of said positional isomers comprises an alpha interferon covalently conjugated to a substantially non-antigenic polymer at a histidine residue on said alpha interferon, wherein said substantially non-antigenic polymer is a polyalkylene oxide comprising an alkyl terminal.

2. The pharmaceutical composition of claim **1**, wherein said alpha interferon is interferon alpha 2b.

3. The pharmaceutical composition of claim **2**, wherein said histidine residue is His34.

4. The pharmaceutical composition of claim **1**, wherein said mixture of said alpha interferon positional isomers comprises at least about 3 positional isomers.

5. The pharmaceutical composition of claim **4**, wherein said mixture of said alpha interferon positional isomers comprises at least about 6 positional isomers.

6. The pharmaceutical composition of claim **5**, wherein said mixture of said alpha interferon positional isomers comprises at least about 8 positional isomers.

7. The pharmaceutical composition of claim **6**, wherein said alpha interferon is alpha interferon 2b and said mixture of positional isomers comprises a substantially non-antigenic polymer linked to said alpha interferon 2b, at an amino acid residue selected from the group consisting of Cys1, Lys31 His34, Lys49, Lys83, Lys121, Lys131 and Lys134.

8. The pharmaceutical composition of claim **1**, wherein said polyalkylene oxide is a polyethylene glycol.

9. The pharmaceutical composition of claim **8**, wherein said polyalkylene oxide is a monomethoxy-polyethylene glycol, (mPEG).

10. The pharmaceutical composition of claim **1**, wherein said substantially non-antigenic polymer has a molecular weight of from about 200 to about 35,000.

11. The pharmaceutical composition of claim **10**, wherein said substantially non-antigenic polymer has a molecular weight of from about 1,000 to about 15,000.

12. The pharmaceutical composition of claim **11**, wherein said substantially non-antigenic polymer has a molecular weight of from about 2,000 to about 12,500.

13. A pharmaceutical composition, comprising a mixture of alpha interferon polymer conjugate positional isomers, wherein one of said positional isomers comprises an alpha interferon covalently conjugated to a substantially non-antigenic polymer at a histidine residue on said alpha interferon, wherein said substantially non-antigenic polymer is selected from the group consisting of polypropylene glycol, dextran, polyvinyl pyrrolidones, polyacryl amides, polyvinyl alcohols and carbohydrate-based polymers.

14. An alpha interferon-containing composition, comprising a plurality of alpha interferon polymer conjugates, wherein at least about 15% of the conjugates include covalent attachment of a substantially non-antigenic polymer at a histidine of said alpha interferon, wherein said substantially non-antigenic polymer is a polyalkylene oxide comprising an alkyl terminal.

15. The composition of claim **14**, wherein the alpha interferon portion of said composition is alpha interferon 2b and said histidine is His34.

16. The composition of claim **14**, wherein at least about 30% of said conjugates include covalent attachment of said substantially non-antigenic polymer at histidine-34 of said alpha interferon.

17. The composition of claim **16**, wherein at least about 40% of said conjugates include covalent attachment of said substantially non-antigenic polymer at histidine-34 of said alpha interferon.

18. A pharmaceutical composition, comprising a mixture of alpha interferon 2b-polymer positional isomers, wherein from about 30 to about 60% of the positional isomers include a substantially non-antigenic polymer conjugated to the His34 of said alpha interferon, from about 7 to about 20% of the positional isomers include a substantially non-antigenic polymer conjugated to the Cys1 of said alpha interferon and about 7 to about 15% of the positional isomers include a substantially non-antigenic polymer conjugated to the Lys121 of said alpha interferon, wherein said substantially non-antigenic polymer is a polyalkylene oxide comprising an alkyl terminal.

19. The pharmaceutical composition of claim **19**, wherein about 55% of the positional isomers include a substantially

non-antigenic polymer conjugated to the His34 of said alpha interferon, about 15% of the positional isomers include a substantially non-antigenic polymer conjugated to the Cys1 of said alpha interferon and about 15% of the positional isomers include a substantially non-antigenic polymer conjugated to the Lys121 of said alpha interferon.

20. A method of preparing alpha-interferon conjugates, comprising contacting an alpha interferon with a sufficient amount of a mono-activated oxycarbonyl-oxy-N-dicarboximide-activated substantially non-antigenic polymer under conditions which are sufficient to facilitate covalent attachment of said substantially non-antigenic polymer at a histidine of said alpha interferon, wherein said substantially non-antigenic polymer is selected from the group consisting of a polyalkylene oxide comprising an alkyl terminal, polypropylene glycol, dextran, polyvinyl pyrrolidones, polyacryl amides, polyvinyl alcohols and carbohydrate-based polymers.

21. The method of claim **20**, wherein said oxycarbonyl-oxy-N-dicarboximide is succinimidyl carbonate.

22. The method of claim **20**, wherein said conditions include conducting said contacting at a pH of less than about 7.0.

23. The method of claim **22**, wherein said conditions include conducting said contacting at a pH of less than about 6.8.

24. The method of claim **23**, wherein said conditions include conducting said contacting at a pH of from about 4.5 to about 6.8.

25. The method of claim **20**, wherein said activated substantially non-antigenic polymer is present in a molar excess with respect to said alpha interferon.

26. The method of claim **20**, wherein said polymer is present in a molar ratio ranging from about 1 to about 8-parts polymer per part alpha interferon.

27. The method of claim **25**, wherein said polymer molar excess is from about 1.5 to about 7-fold.

28. The method of claim **17**, wherein said polymer molar excess is about 1.75 to about 5-fold.

29. The method of claim **20**, wherein said polyalkylene oxide is a polyethylene glycol.

30. The method of claim **20**, wherein said substantially non-antigenic polymer has a molecular weight of from about 200 to about 35,000.

31. The method of claim **30**, wherein said substantially non-antigenic polymer has a molecular weight of from about 1,000 to about 15,000.

32. The method of claim **31**, wherein said substantially non-antigenic polymer has a molecular weight of from about 2,000 to about 12,500.

33. The method of claim **20**, wherein said alpha interferon is interferon alpha 2b.

34. A method of treating an interferon-susceptible condition in mammals, comprising administering an effective amount of a composition of claim **1**.

35. A method of treating an interferon-susceptible condition in mammals, comprising administering an effective amount of a composition of claim **14**.

36. A method of treating an interferon-susceptible condition in mammals, comprising administering an effective amount of a composition of claim **15**.

37. A substantially non-antigenic polymer-interferon conjugate prepared according to the method of claim **20**.

38. The pharmaceutical composition of claim **13**, wherein said alpha interferon is interferon alpha 2b.

39. The pharmaceutical composition of claim **38**, wherein said histidine residue is His34.

40. The pharmaceutical composition of claim **13**, wherein said mixture of said alpha interferon positional isomers comprises at least about 3 positional isomers.

41. The pharmaceutical composition of claim **13**, wherein said mixture of said alpha interferon positional isomers comprises at least about 6 positional isomers.

42. The pharmaceutical composition of claim **13**, wherein said mixture of said alpha interferon positional isomers comprises at least about 8 positional isomers.

43. The pharmaceutical composition of claim **38**, wherein said mixture of positional isomers comprises a substantially non-antigenic polymer linked to said alpha interferon 2b, at an amino acid residue selected from the group consisting of Cys1, Lys31, His34, Lys49, Lys83, Lys121, Lys131 and Lys134.

44. The pharmaceutical composition of claim **13**, wherein said substantially non-antigenic polymer has a molecular weight of from about 200 to about 35,000.

45. The pharmaceutical composition of claim **13**, wherein said substantially non-antigenic polymer has a molecular weight of from about 1,000 to about 15,000.

46. The pharmaceutical composition of claim **13**, wherein said substantially non-antigenic polymer has a molecular weight of from about 2,000 to about 12,500.

47. The pharmaceutical composition of claim **1** wherein said polyalkylene oxide is terminated with a C_{1-4} alkyl.

48. The pharmaceutical composition of claim **18** wherein said polyalkylene oxide is terminated with a C_{1-4} alkyl.

49. The method of claim **20** wherein said polyalkylene oxide is terminated with a C_{1-4} alkyl.

50. A pharmaceutical composition, comprising a mixture of alpha interferon polymer conjugate positional isomers, wherein at least one of said positional isomers comprises an alpha interferon covalently conjugated to a substantially non-antigenic polymer at a histidine residue on said alpha interferon, prepared by a process comprising contacting an alpha interferon with a sufficient amount of a mono-activated substantially non-antigenic polymer under conditions which are sufficient to facilitate covalent attachment of said substantially non-antigenic polymer at a histidine of said alpha interferon, said mono-activated substantially non-antigenic polymer being a polyalkylene oxide comprising an alkyl substituted terminal.

* * * * *

SCHERING CORPORATION

EXHIBIT IV

GALLOPING HILL ROAD KENILWORTH, N.J. 07033

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0005

June 2, 1997

Food and Drug Administration
Center for Biologics Evaluation and Research
Document Control Center (HFM-99)
Woodmont Office Center, Suite 200N
1401 Rockville Pike
Rockville, Maryland 20852-1448
Attn: Ms. Sharon Risso, Director
Office of Therapeutics Research and Review
Division of Application Review and Policy (HFM-585)

SCH 54031
(PEG_{12000}-Interferon)
alfa-2b
Serial No. 000

SUBJECT: INVESTIGATIONAL NEW DRUG APPLICATION

Dear Ms. Risso:

Enclosed herein please find our Investigational New Drug Application for our Pegylated Interferon alfa-2b (SCH 54031).

We acknowledged your Pre-IND comments from the meeting held on May 13, 1997 to assist us in further development of this compound.

Changes were made to our protocol C97-010 entitled "Comparison of PEG-IFN vs. Intron A for Treatment of Adult Subjects with Chronic Hepatitis C not Previously Treated with Interferon: Dose Finding Study" that was submitted on May 2, 1997 prior to the Pre-IND meeting. Per your suggestions we have included ALTs' and HCV-RNA as the primary endpoint and we will use conventional post-treatment follow-up data to support registration in the U.S.

As discussed with Ms. Gress on May 22, 1997 the protocol still has an interim analysis. Results from this analysis may be used to support international findings prior to the BLA filing in the U.S.

Please be advised that material and data contained in this submission are considered to be confidential. The legal protection of such confidential commercial material is claimed under the applicable provisions of 18 U.S.C., Section 1905 or 21 U.S.C., Section 331(j) as well as the FDA regulations.

Sincerely,

Malvina Landrigan for
Tobias Massa, Ph.D.
Senior Director, Technical Support
Worldwide Regulatory Affairs

ML/pjm
Enclosures

EXHIBIT V

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
1401 Rockville Pike
Rockville MD 20852-1448

RECEIVED
JUN 10 1997
TOBIAS MASSA, Ph.D.

JUN - 5 1997

Our Reference: BB-IND 7173

Schering Corporation
Attention: Tobias Massa, Ph.D.
Senior Director, Technical Support
Worldwide Regulatory Affairs
2000 Galloping Hill Road
Kenilworth, NJ 07033

Dear Dr. Massa:

The Center for Biologics Evaluation and Research has received your **Investigational New Drug Application (IND)**. The following product name and BB-IND number have been assigned to this application. They serve only to identify it and do not imply that this Center either endorses or does not endorse your application.

BB-IND #: **7173**

SPONSOR: **Schering Corporation**

PRODUCT NAME: **Pegylated Interferon Alfa-2b (human, recombinant, *E.coli*, Schering)**

DATE OF SUBMISSION: **June 2, 1997**

DATE OF RECEIPT: **June 3, 1997**

This BB-IND number should be used to identify all future correspondence and submissions, as well as telephone inquiries concerning this IND. Please provide an **original and two copies of every submission to this file**. Please include three originals of all illustrations which do not reproduce well.

It is understood that studies in humans will not be initiated until 30 days after the date of receipt shown above. If this office notifies you, verbally or in writing, of serious deficiencies that require correction before human studies can begin, it is understood that you will continue to withhold such studies until you are notified that the material you have submitted to correct the deficiencies is satisfactory. If such a clinical hold is placed on this file, you will be notified in writing of the reasons for placing the IND on hold.

You are responsible for compliance with applicable portions of the Public Health Service Act, the Federal Food, Drug, and Cosmetic Act, and the Code of Federal Regulations (CFR). A

copy of 21 CFR Part 312, pertaining to INDs, is enclosed. Copies of other pertinent regulations are available from this Center upon request. The following points regarding obligations of an IND sponsor are included for your information only, and are not intended to be comprehensive.

Progress reports are required at intervals not exceeding one year and are due within 60 days of the anniversary of the date that the IND went into effect. Any unexpected, fatal or immediately life-threatening reaction which is associated with use of this product must be reported to this Center within three working days, and all serious, unexpected adverse experiences must be reported, in writing, to this Center and to all study centers within ten working days.

Charging for an investigational product in a clinical trial under an IND is not permitted without the prior written approval of the FDA.

Prior to use of each new lot of the investigational biologic in clinical trials, please submit the lot number, the results of all tests performed on the lot, and the specifications when established (i.e., the range of acceptable results).

If not included in your submission, please provide copies of the consent forms for each clinical study. A copy of the requirements for and elements of informed consent are enclosed. Also, please provide documentation of the institutional review board approval(s) for each clinical study.

All laboratory or animal studies intended to support the safety of this product should be conducted in compliance with the regulations for "Good Laboratory Practice for Nonclinical Laboratory Studies" (21 CFR Part 58, copies available upon request). If such studies have not been conducted in compliance with these regulations, please provide a statement describing in detail all differences between the practices used and those required in the regulations.

Item 7a of form FDA 1571 requests that either an "environmental assessment," or a "claim for categorical exclusion" from the requirements for environmental assessment, be included in the IND. If you did not include a response to this item with your application, please submit one. See the enclosed information sheet for additional information on how these requirements may be addressed.

Sponsors of INDs for products used to treat life-threatening or severely debilitating diseases are encouraged to consider the interim rule outlined in 21 CFR 312.80 through 312.88.

Telephone inquiries concerning this IND should be made directly to me at (301) 827-5101. Correspondence regarding this file should be addressed as follows:

Center for Biologics Evaluation and Research
Attn: Office of Therapeutics Research and Review
HFM-99, Room 200N
1401 Rockville Pike
Rockville, MD 20852-1448

If we have any comments after we have reviewed this submission, we will contact you.

Sincerely yours,

Victoria Tyson-Medlock

Victoria Tyson-Medlock
Consumer Safety Officer
Division of Application Review and Policy
Office of Therapeutics
Research and Review
Center for Biologics
Evaluation and Research

Enclosures (3): 21 CFR Part 312
21 CFR 50.20, 50.25
Information sheet on 21 CFR 25.24

EXHIBIT VI

SCHERING CORPORATION

2000 GALLOPING HILL ROAD KENILWORTH, N.J. 07033

TELEPHONE: (908) 298-4000

December 22, 1999

Food and Drug Administration
Center for Biologics Evaluation and Research
Office of Therapeutics Research and Review
Division of Application Review and Policy (HFM-585)
c/o Document Control Center (HFM-99)
12100 Parklawn Drive, 1st Floor
Rockville, Maryland 20852

BLA
PEG-Intron™
(Peginterferon alfa-2b)
SCH 54031

SUBJECT: ORIGINAL BIOLOGICS LICENSE APPLICATION

Dear Dr. Jones:

Schering Corporation is submitting an original Biologics License Application (BLA) for PEG-Intron™, peginterferon alfa-2b, therapy for the treatment of chronic hepatitis C.

Request and Justification for Priority Review. Schering is hereby requesting a priority review for this application.

Hepatitis C virus (HCV) infection is a major cause of chronic liver disease and is a significant health–care problem throughout the world. The natural history of HCV infection appears to be that of a silent but progressive chronic inflammation of the liver. Chronic disease develops in 60–80% of patients and 20–50% of these develop cirrhosis with the accompanying risk of liver failure and hepatocellular carcinoma. Chronic hepatitis C (CHC) is the most frequent underlying condition necessitating liver transplantation in the US.

In the US, it is estimated that 1.8% of the population is chronically infected. This means that chronic hepatitis C currently affects an estimated 3.9 million Americans. The CDC estimates that HCV infections contribute to approximately 12,000 deaths each year, with the number expected to triple by the year 2010. The costs of this disease to the health care system are in the range of $500 million annually.

At the time the pivotal clinical trial with PEG-Intron was initiated, the only approved therapies for CHC were the alpha interferons. These treatments were recognized to be suboptimal due to a relatively low initial response and high relapse rate which results in sustained response in only approximately 12% of patients. In addition, these treatments require thrice weekly injections and pose a considerable burden on the patients.

First line therapy for CHC has since evolved to REBETRON™ Combination Therapy with Intron A (interferon alfa-2b) for Injection and Rebetol® (ribavirin) capsules. This therapy results in a substantial increase in the number of patients who remain free of detectable HCV-RNA after treatment cessation (sustained virologic response of approximately 40% in treatment naïve patients).

It is recognized that, currently, REBETRON Combination Therapy provides the best efficacy and should be considered first line therapy for most patients. However, there remains a need for alternate treatments which provide useful therapeutic options for patients who may be at an unacceptable risk with using ribavirin or who cannot tolerate the REBETRON Combination Therapy. The Rebetol capsules cause additional side effects and risks which are not associated with interferon based monotherapies, including PEG-Intron. These additional side effects include hemolysis, teratogenicity, dyspnea and pruritis, which may be unacceptable for some patients.

In addition, some patients have difficulty in continuing thrice weekly injections for the entire 48 weeks of recommended therapy. For these patients PEG-Intron is an alternate therapy requiring only once weekly injections. As an alternate treatment, PEG-Intron provides efficacy better than the only other alternatives currently available (i.e. alpha interferons) and in a subset of patients, efficacy comparable to Rebetron can be achieved.

Among patients with low baseline viral load ($\leq 2 \times 10^6$ copies/ml), PEG-Intron, at 1.0 µg/kg, results in a sustained response rate of 47%. In separate clinical trials in which Rebetron Combination therapy was tested, the same subgroup of patients with baseline HCV-RNA $\leq 2 \times 10^6$, the combination produced an observed sustained response rate of 46%. Thus, for this subgroup, PEG-Intron is a viable therapeutic alternative with comparable efficacy and a potential safety advantage (with respect to ribavirin specific side effects). The once weekly injection schedule is also an advantage likely to enhance compliance. Thus, PEG-Intron can become the alternative choice to REBETRON Combination Therapy.

In accordance with CBER's criteria, new therapies for serious diseases may be assigned for Priority review, not only for improvement in efficacy over available therapies but also for products which improve compliance or eliminate or reduce treatment-limiting drug reactions. With the once weekly regimen recommended for

PEG-Intron administration, the potential for patient compliance is enhanced and, as noted above, PEG-Intron does not cause certain potentially treatment-limiting side effects associated with ribavirin.

Chronically infected hepatitis C patients represent a large group with a significant medical need. The results of the pivotal clinical study confirms the benefit of treatment with PEG-Intron compared with the only other therapy, e.g. alpha interferon monotherapy available as an alternative to REBETRON Combination Therapy with Intron A and Rebetol.

As detailed in the application, it is the opinion of Schering–Plough Corporation that treatment with the PEG-Intron at the recommended target dose of 1 µg/kg QW provides a new therapeutic tool in the treatment of chronic hepatitis C by significantly improving the patient's ability to achieve sustained response to treatment relative to Intron A. This is particularly important for patients for whom the risk/benefit assessment for using ribavirin is unfavorable.

CALA. This submission is also being submitted as a Computer Assisted License Application (CALA). The CALA consists of a clinical data section (Business Objects), and read-only electronically published documents (PDF). Please note that SAS data files and programs are provided in Section 11 of the application and an additional set of this information is provided on CD-ROM for the statistical reviewer.

Pre-BLA Discussions. The October 1999 Briefing Book containing the efficacy and safety data from Schering's large pivotal trial was provided to the Agency on October 6, 1999. The Pre-BLA meeting was held on November 2, 1999. At this meeting, Schering presented the following questions and agreements are noted below:

Fileability. The Agency found the proposed BLA acceptable for filing with caveats (single study, relatively small numbers at the proposed dose for such a prevalent disease). However, it was agreed that these are primarily review issues (i.e. approvability vs. 'fileability').

ISS/ISE. FDA agreed with our proposal that an ISS and ISE would not add to the application and hence were not necessary for the submission.

Priority Review. Priority review of the PEG-Intron monotherapy BLA was discussed. It was noted that PEG-Intron might offer a therapeutic option for patients unable tolerate Rebetron. It is recognized that this specific subgroup was not studied.

Co-Primary Endpoint for U.S. and European Applications. FDA requested that Schering provide a review with supporting documentation of historical discussions between SPRI and FDA regarding the primary endpoint used in the pivotal study. This review was submitted November 29, 1999 (IND 7173, Serial No. 162).

Pediatric Studies. A waiver for PEG-Intron monotherapy studies might not be applicable as there is a potential concern about the toxicity of Rebetron in children. SPRI agreed to discuss with the HCV community regarding potential utility of PEG monotherapy in pediatric treatment. A deferral of pediatric data was acceptable. A pediatric study deferral request was submitted to the Agency on September 1, 1999.

Preclinical Reproductive Studies. As requested by the Agency during a teleconference on April 22, 1999, Schering is performing a study to demonstrate that PEG-Intron, like other interferons, affects estrogen and progesterone. Schering is evaluating the progesterone and estradiol levels in cynomolgus monkeys as a surrogate for the reproductive toxicities of PEG-Intron. This study design was discussed on several occasions with Dr. Anne Pilaro of CBER and comments received in FDA's September 24, 1999 letter were incorporated. Schering and the Agency agreed that this study report would be provided during the review cycle of the application.

CMC. In a pre-BLA teleconference with Drs. Dye, Bekisz and Nguyen, CBER on October 20, 1999, the following issues were discussed and agreed upon:

- Peginterferon alfa-2b meets the definition of a specified (i.e. well-characterized) biologic. As such, the CMC section of the BLA conforms with the FDA "Guidance for Submission of the CMC Information for Therapeutic Recombinant DNA-Derived Products or Monoclonal Antibody Products for In Vivo Use".
- Stability data presented in the Briefing Book supports a shelf-life recommendation of 24 months at 25°C for PEG-Intron vial strengths between 50 and 150 µg.
- Filing an amendment for a new vial strength (35 µg) on or before June 2000 would not impact a standard review clock for the BLA.
- An explanation would be provided in BLA for the high endotoxin result on one of the peginterferon drug substance batches. In a follow-up call with Dr. Bekisz on November 2, it was noted that the high endotoxin result on this batch was within specification (≤ 75 EU/ml) at the time of release. We agreed to identify in the BLA which drug substance batches were released based on the ≤ 75 EU/ml specification.
- A rationale for deleting the SDS-PAGE purity test for drug substance and drug product would be provided in the BLA.

Please note that throughout this application, PEG-Intron is also referred to as PEG-IFN, peginterferon alfa-2b, peginterferon, and SCH 54031. These names all refer to the same biological product.

Debarment Certification. In accordance with Section 306(k) of the Food, Drug and Cosmetic Act, Schering Corporation certifies that, with respect to this application, it did not and will not knowingly use the services of any persons that have been debarred under the provisions of Section 306(a) or (b) of the Act.

A check in the amount of $272,282.00 was sent to the FDA c/o Mellon Bank, Pittsburgh, PA on December 22, 1999. This check represents the 1999 application fee for this BLA. The User Fee ID Number is 0994KA1414Dec99.

Please be advised that material and data contained in this submission are considered to be confidential. The legal protection of such confidential commercial material is claimed under the applicable provisions of 18 U.S.C., Section 1905 or 21 U.S.C., Section 331(j) as well as the FDA regulations.

Sincerely,

Nicholas J. Pelliccione, Ph.D.
Senior Director, Technical Support
Worldwide Regulatory Affairs

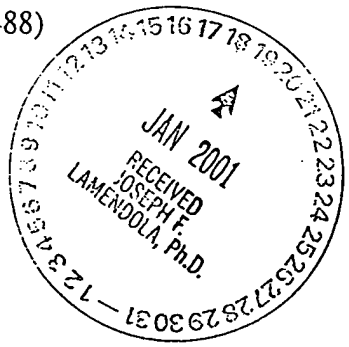
RAS/kw
Enclosures

DEPARTMENT OF HEALTH & HUMAN SERVICES

EXHIBIT VII

Food and Drug Administration
1401 Rockville Pike
Rockville MD 20852-1448

Our STN: BL 103949 (replaces Ref. No. 99-1488)



January 19, 2001

Nicholas J. Pelliccione, Ph.D.
Schering Corporation
2000 Galloping Hill Road
Kenilworth, NJ 07033

Dear Dr. Pelliccione:

Your biologics license application for Peginterferon alfa-2b is approved effective this date. Schering Corporation, Kenilworth, New Jersey, is hereby authorized to introduce or deliver for introduction into interstate commerce, Peginterferon alfa-2b under Department of Health and Human Services U.S. License No. 0994.

Peginterferon alfa-2b is indicated for the treatment of chronic hepatitis C in patients not previously treated with interferon alfa who have compensated liver disease and are at least 18 years of age. Under this authorization, you are approved to manufacture Peginterferon alfa-2b at your facility in Innishannon County Cork, Ireland. Final formulated drug product will be filled at Innishannon County Cork and unlabeled vials of drug product will be shipped to Kenilworth, New Jersey, for labeling, packaging and distribution. In accordance with approved labeling, your product will bear the trade name PEG-Intron, and will be marketed in 100 µg /mL, 160 µg /mL, 240 µg /mL and 300 µg /mL vials of lyophilized powder, supplied with a 5-mL vial of PEG-Intron Diluent (Sterile Water for Injection), two disposable 1-mL (Becton Dickenson Safety-Lok) syringes with needles and needle guards, and alcohol swabs.

The dating period for Peginterferon alfa-2b shall be 24 months from the date of manufacture when stored at 25 °C (77 ° F). The date of manufacture shall be defined as the date of final sterile filtration of the formulated drug product. The bulk drug substance may be stored for up to 36 months at -80 °C. Results of ongoing stability studies should be submitted throughout the dating period, as they become available, including the results of stability studies from the first three production lots. The stability protocol in your license application is considered approved for the purpose of extending the expiration dating period of your drug substance and drug product as specified in 21 CFR 601.12.

You are not currently required to submit samples of future lots of Peginterferon alfa-2b to the Center for Biologics Evaluation and Research (CBER) for release by the Director, CBER, under 21 CFR 610.2. FDA will continue to monitor compliance with 21 CFR 610.1 requiring assay and release of only those lots that meet release specifications.

Any changes in the manufacturing, testing, packaging or labeling of Peginterferon alfa-2b, or in the manufacturing facilities will require the submission of information to your biologics license application for our review and written approval consistent with 21 CFR 601.12.

Any changes in the manufacturing, testing, packaging or labeling of Peginterferon alfa-2b, or in the manufacturing facilities will require the submission of information to your biologics license application for our review and written approval consistent with 21 CFR 601.12.

As of April 1, 1999, all applications for new active ingredients, new dosage forms, new indications, new routes of administration, and new dosing regimens are required to contain an assessment of safety and effectiveness of the product in pediatric patients unless this requirement is waived or deferred (63 FR 66632). We do not concur with your request, as submitted to your application on February 4, 2000, to waive the requirement to conduct pediatric studies. As communicated during the December 14, 2000, meeting, we are deferring the submission of your pediatric studies until June 30, 2001, subsequent to discussion at an open session of an FDA advisory committee meeting.

Pursuant to 21 CFR Part 208, FDA has determined that this product poses a serious and significant public health concern requiring the distribution of a Medication Guide. Distribution of a Medication Guide is necessary for safe and effective use of this product. FDA has determined that Peginterferon alfa-2b is a product for which patient labeling could help prevent serious adverse effects and inform the patient of serious risks relative to benefit that could affect their decisions to use, or continue to use the product. *See* 21 CFR 208.1. FDA hereby approves the Medication Guide you submitted January 19, 2001. In accordance with 21 CFR 208, you are responsible for ensuring that this Medication Guide is available for every patient who is dispensed a prescription for this product. In addition, you are responsible for ensuring that the label of each package includes a prominent and conspicuous instruction to authorized dispensers to provide a Medication Guide to each patient to whom the drug is dispensed, and states how the Medication Guide is provided.

We acknowledge your written commitments to provide additional information and to conduct post marketing studies as described in your letters of November 28, 2000, and January 12, 2001, as outlined below:

1. To address the safety and efficacy of Peginterferon alfa-2b in African Americans by submitting the data from a study of 100 previously untreated patients with chronic hepatitis C who will receive 1.5 µg/kg PEG-Intron and 800, 1000 or 1200 mg ribavirin, depending on their weight. The final protocol for this study will be submitted to CBER by April 1, 2001. Patient accrual will be completed by June 1, 2002, the study completed by December 1, 2003 and a final study report submitted to CBER by June 1, 2004.

2. To evaluate, in patients diagnosed with chronic hepatitis C and compensated liver disease, the effects of single and multiple doses of Peginterferon alfa-2b on the disposition of drugs known to be metabolized by hepatic cytochrome P450 enzymes. The final protocol for this study will be submitted to CBER by February 22, 2001. Patient accrual will be completed by February 19, 2002, the study completed by April 19, 2002, and a final study report submitted to CBER by November 20, 2002.

3. To evaluate the pharmacokinetic, pharmacodynamic and clinical effects of Peginterferon alfa-2b when given chronically to patients with renal dysfunction (creatinine clearance <50 mL/min). The final protocol for this study will be submitted to CBER by March 1, 2001. Patient accrual will be completed by March 1, 2002, the study completed by April 29, 2002, and a final study report submitted to CBER by October 21, 2002.

4. To evaluate the pharmacokinetic, pharmacodynamic and clinical effects of Peginterferon alfa-2b when administered to patients receiving methadone. The final protocol for this study will be submitted to CBER by May 15, 2001. Patient accrual will be completed by May 12, 2004, the study completed by July 12, 2004, and a final study report submitted to CBER by January 18, 2005.

5. To replace the 5-mL vial of diluent that is packaged with Peginterferon alfa-2b with a 1-mL vial of diluent. The supplement supporting this change will be submitted by December 31, 2001.

It is requested that adverse experience reports be submitted in accordance with the adverse experience reporting requirements for licensed biological products (21 CFR 600.80) and that distribution reports be submitted as described (21 CFR 600.81). All adverse experience reports should be prominently identified according to 21 CFR 600.80 and be submitted to the Center for Biologics Evaluation and Research, HFM-210, Food and Drug Administration, 1401 Rockville Pike, Rockville, MD 20852-1448.

Please submit all final printed labeling at the time of use and include implementation information on FDA Form 2567. Please provide a PDF-format electronic copy as well as original paper copies (ten for circulars and five for other labels). In addition, you may wish to submit draft copies of the proposed introductory advertising and promotional labeling with an FDA Form 2567 or Form 2253 to the Center for Biologics Evaluation and Research, Advertising and Promotional Labeling Branch, HFM-602, 1401 Rockville Pike, Rockville, MD 20852-1448. Final printed advertising and promotional labeling should be submitted at the time of initial dissemination, accompanied by a FDA Form 2567 or Form 2253.

All promotional claims must be consistent with and not contrary to approved labeling. No comparative promotional claim or claim of superiority over other products should be made unless data to support such claims are submitted to and approved by the Center for Biologics Evaluation and Research.

Sincerely yours,

Shawn L Rosso

for Jay P. Siegel, M.D., FACP
Director
Office of Therapeutics
Research and Review
Center for Biologics
Evaluation and Research

UNITED STATES DEPARTMENT OF COMMERCE
Patent and Trademark Office
ASSISTANT SECRETARY AND COMMISSIONER
OF PATENTS AND TRADEMARKS
Washington, D.C. 20231

EXHIBIT VIII

MAY 28, 1998

PTAS

100662289A

MICHAEL N. MERCANTI
P.O. BOX 484
PRINCETON, NJ 08542

UNITED STATES PATENT AND TRADEMARK OFFICE
NOTICE OF RECORDATION OF ASSIGNMENT DOCUMENT

THE ENCLOSED DOCUMENT HAS BEEN RECORDED BY THE ASSIGNMENT DIVISION OF THE U.S. PATENT AND TRADEMARK OFFICE. A COMPLETE MICROFILM COPY IS AVAILABLE AT THE ASSIGNMENT SEARCH ROOM ON THE REEL AND FRAME NUMBER REFERENCED BELOW.

PLEASE REVIEW ALL INFORMATION CONTAINED ON THIS NOTICE. THE INFORMATION CONTAINED ON THIS RECORDATION NOTICE REFLECTS THE DATA PRESENT IN THE PATENT AND TRADEMARK ASSIGNMENT SYSTEM. IF YOU SHOULD FIND ANY ERRORS OR HAVE QUESTIONS CONCERNING THIS NOTICE, YOU MAY CONTACT THE EMPLOYEE WHOSE NAME APPEARS ON THIS NOTICE AT 703-308-9723. PLEASE SEND REQUEST FOR CORRECTION TO: U.S. PATENT AND TRADEMARK OFFICE, ASSIGNMENT DIVISION, BOX ASSIGNMENTS, CG-4, 1213 JEFFERSON DAVIS HWY, SUITE 320, WASHINGTON, D.C. 20231.

RECORDATION DATE: 12/22/1997

REEL/FRAME: 9063/0929
NUMBER OF PAGES: 3

BRIEF: ASSIGNMENT OF ASSIGNOR'S INTEREST (SEE DOCUMENT FOR DETAILS).

ASSIGNOR:
PARK-CHO, MYUNG-OK

DOC DATE: 11/11/1997

ASSIGNEE:
ENZON, INC.
20 KINGSBRIDGE ROAD
PISCATAWAY, NEW JERSEY 08854

SERIAL NUMBER: 08994622
PATENT NUMBER:

FILING DATE: 12/19/1997
ISSUE DATE:

MAYA BENNETT, EXAMINER
ASSIGNMENT DIVISION
OFFICE OF PUBLIC RECORDS

RESUB

U.S. DEPARTMENT OF COMMERCE
Patent and Trademark Office

FORM PTO-1595 (Modified)
(Rev. 6-93)
OMB No. 0651-0011 (exp.4/94)
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P08A/REV02 MRD 12-22-97

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03-16-1998

100662289

DEC 22 1997

3/6/98

ied original documents or copy thereof.

To the Honorable Commissioner of Patents

1. Name of conveying party(ies):
Myung-ok PARK-CHO

Additional names(s) of conveying party(ies) attached? ☐ Yes ☒ No

2. Name and address of receiving party(ies):

Name: Enzon, Inc.

Address: 20 Kingsbridge Road

City: Piscataway State/Prov.: NJ

Country: USA ZIP: 08854

Additional name(s) & address(es) attached? ☐ Yes ☒ No

08/994622

3. Nature of conveyance:

☒ Assignment ☐ Merger

☐ Security Agreement ☐ Change of Name

☐ Other _____

Execution Date: _____

4. Application number(s) or registration numbers(s):

If this document is being filed together with a new application, the execution date of the application is: November 11, 1997

Patent Application No. Filing date

B. Patent No.(s)

Additional numbers attached? ☐ Yes ☒ No

5. Name and address of party to whom correspondence concerning document should be mailed:

Name: Michael N. Mercanti

Registration No. 33,966

Address: P.O. Box 484

City: Princeton State/Prov.: NJ

Country: USA ZIP: 08542

6. Total number of applications and patents involved: 1

7. Total fee (37 CFR 3.41):.....$ 40.00

☒ Enclosed

☐ Authorized to be charged to deposit account

8. Deposit account number:

DO NOT USE THIS SPACE

01/07/1998 PARK-CHO M 00000264
40.00 OP
01 FC:581

9. Statement and signature.

To the best of my knowledge and belief, the foregoing information is true and correct and any attached copy is a true cc of the original document.

Michael N. Mercanti — Name of Person Signing

Signature

December 9, 1997 — Date

Total number of pages including cover sheet, attachments, and document: 3

Docket No.: 213.1020CIP2

ASSIGNMENT

WHEREAS, I (we), Carl W. Gilbert and Myung-ok Park-Cho, residing at 4655 Oakleigh Manor Drive, Powder Springs, GA 30127 and 1-207 Dong, A Apt., Chang-Dong, Tobong-Gu, Seoul, Korea, respectively, have invented certain new and useful improvements in IMPROVED INTERFERON POLYMER CONJUGATES for which I (we) are about to execute an application for a Patent of the United States

X which is executed on November 11, 1997
X which is identified by ROBERTS & MERCANTI, LLP, Docket No. 213.1020CIP2
_ which is filed on _____, Serial No. _____
X I (we) hereby authorize and request my (our) attorney, Roberts & Mercanti, LLP of P.O. Box 484 Princeton, New Jersey 08542-0484 to insert here in parentheses (Application number _____, filed _____) the filing date and application number of said application when known.

and WHEREAS, Enzon, Inc., 20 Kingsbridge Road, Piscataway, NJ 08854-3998, ASSIGNEE is desirous of obtaining the entire right, title and interest in, to and under the said invention and the said application:

NOW, THEREFORE, in consideration of the sum of One Dollar ($1.00) to me (us) in hand paid, and other good and valuable consideration, the receipt of which is hereby acknowledged, I (we), the said ASSIGNOR(S), have sold, assigned, transferred and set over, and by these presents do hereby sell, assign, transfer and set over, unto the said ASSIGNEE, its successors, legal representatives and assigns, the entire right, title and interest in, to and under the said invention, and the said United States application and all divisions, renewals and continuations thereof, and all Patents of the United States which may be granted thereon and all reissues and extensions thereof; and all applications for industrial property protection, including, without limitation, all applications for patents, utility models, and designs which may hereafter be filed for said invention in any country or countries foreign to the United States, together with the right to file such applications and the right to claim for the same the priority rights derived from said United States application under the Patent Laws of the United States, the International Convention for the Protection of Industrial Property, or any other international agreement or the domestic laws of the country in which any such application is filed, as may be applicable; and all forms of industrial property protection, including, without limitation, patents, utility models, inventors' certificates and designs which may be granted for said invention in any country or countries foreign to the United States and all extensions, renewals and reissues thereof;

AND I (WE) HEREBY authorize and request the Commissioner of Patents and Trademarks of the United States, and any Official of any country or countries foreign to the United States, whose duty it is to issue patents or other evidence or forms of industrial property protection on applications as aforesaid, to issue the same to the said ASSIGNEE, its successors, legal representatives and assigns, in accordance with the terms of this instrument.

AND I (WE) HEREBY covenant and agree that I (we) have full right to convey the entire interest herein assigned, and that I (we) have not executed, and will not execute, any agreement in conflict herewith.

AND I (WE) HEREBY further covenant and agree that I (we) will communicate to the said ASSIGNEE, its successors, legal representatives and assigns, any facts known to me (us) respecting said invention, and testify in any legal proceeding, sign all lawful papers, execute all divisional, continuing, reissue and foreign applications, make all rightful oaths, and generally do everything possible to aid the said ASSIGNEE, its successors, legal representatives and assigns, to obtain and enforce proper protection for said invention in all countries.

IN TESTIMONY WHEREOF, I (we) hereunto set my (our) hand and seal the day and year set opposite my (our) signature(s).

Date_____, 1997

_____
(Signature of Inventor)

Carl W. Gilbert
(Typed Name of Inventor)

Date November 11, 1997

x M.P. Cho
(Signature of Co-inventor)

Myung-ok Park-Cho
(Typed Name of Co-inventor)

A:\assignment.wpd

EXHIBIT IX

UNITED ST... DEPARTMENT OF COMMERCE
Patent and Trademark Office
ASSISTANT SECRETARY AND COMMISSIONER
OF PATENTS AND TRADEMARKS
Washington, D.C. 20231

MAY 28, 1998

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100662290A

MICHAEL N. MERCANTI
P.O. BOX 484
PRINCETON, NJ 08542

UNITED STATES PATENT AND TRADEMARK OFFICE
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RECORDATION DATE: 03/06/1998

REEL/FRAME: 9063/0961
NUMBER OF PAGES: 3

BRIEF: ASSIGNMENT OF ASSIGNOR'S INTEREST (SEE DOCUMENT FOR DETAILS).

ASSIGNOR:
GILBERT, CARL W.

DOC DATE: 11/07/1997

ASSIGNEE:
ENZON, INC.
20 KINGSBRIDGE ROAD
PISCATAWAY, NEW JERSEY 08854

SERIAL NUMBER: 08994622
PATENT NUMBER:

FILING DATE: 12/19/1997
ISSUE DATE:

STEVEN POST, EXAMINER
ASSIGNMENT DIVISION
OFFICE OF PUBLIC RECORDS

Docket No.: 213.1020CIP2

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(Rev. 6-93)
OMB No. 0651-0011 (exp.4/94)
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03-16-1998

100662290

SHEET R

U.S. DEPARTMENT OF COMMERCE
Patent and Trademark Office

3/16/98

To the Honorable Commissioner of [illegible] the attached original documents or copy thereof.

1. Name of conveying party(ies):
Carl W. GILBERT

Additional names(s) of conveying party(ies) attached? ☐ Yes ☒ No

2. Name and address of receiving party(ies):

Name: Enzon, Inc.

Address: 20 Kingsbridge Road

City: Piscataway State/Prov.: NJ

Country: USA ZIP: 08854

Additional name(s) & address(es) attached? ☐ Yes ☒ No

3. Nature of conveyance:

☒ Assignment ☐ Merger

☐ Security Agreement ☐ Change of Name

☐ Other _____

Execution Date: November 7, 1997

4. Application number(s) or registration numbers(s):

If this document is being filed together with a new application, the execution date of the application is: November 7, 1997

Patent Application No.	Filing date	B. Patent No.(s)
08/994,622	December 19, 1997	

Additional numbers attached? ☐ Yes ☒ No

5. Name and address of party to whom correspondence concerning document should be mailed:

Name: Michael N. Mercanti

Registration No. 33,966

Address: P.O. Box 484

City: Princeton State/Prov.: NJ

Country: USA ZIP: 08542

6. Total number of applications and patents involved: 1

7. Total fee (37 CFR 3.41):.....$ 40.00

Previously
☒ Enclosed

☐ Authorized to be charged to deposit account

8. Deposit account number:

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9. Statement and signature.

To the best of my knowledge and belief, the foregoing information is true and correct and any attached copy is a true copy of the original document.

Michael N. Mercanti — Name of Person Signing

[signature] — Signature

March 4, 1998 — Date

Total number of pages including cover sheet, attachments, and document: 3

Docket No.: 213.1020CIP2

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To the Honorable Commissioner of Patents and Trademarks: Please record the attached original documents or copy thereof.

1. Name of conveying party(ies):
Carl W. GILBERT

Additional names(s) of conveying party(ies) attached? ☐ Yes ☒ No

2. Name and address of receiving party(ies):

Name: Enzon, Inc.

Address: 20 Kingsbridge Road

City: Piscataway State/Prov.: NJ

Country: USA ZIP: 08854

Additional name(s) & address(es) attached? ☐ Yes ☒ No

3. Nature of conveyance:

☒ Assignment ☐ Merger

☐ Security Agreement ☐ Change of Name

☐ Other

Execution Date:

4. Application number(s) or registration numbers(s):

If this document is being filed together with a new application, the execution date of the application is: November 7, 1997

Patent Application No.	Filing date	B. Patent No.(s)

Additional numbers attached? ☐ Yes ☒ No

5. Name and address of party to whom correspondence concerning document should be mailed:

Name: Michael N. Mercanti

Registration No. 33,966

Address: P.O. Box 484

City: Princeton State/Prov.: NJ

Country: USA ZIP: 08542

6. Total number of applications and patents involved: 1

7. Total fee (37 CFR 3.41):.....$ 40.00

☒ Enclosed

☐ Authorized to be charged to deposit account

8. Deposit account number:

000000265 GILBERT C
40.00 OP

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9. Statement and signature.

To the best of my knowledge and belief, the foregoing information is true and correct and any attached copy is a true copy of the original document.

Michael N. Mercanti — Name of Person Signing

Signature

December 19, 1997 — Date

Total number of pages including cover sheet, attachments, and document:

04/07/1998 DTDW1 01 FC:581

Docket No.: 213.1020CIP2

ASSIGNMENT

WHEREAS, I (we), Carl W. Gilbert and Myung-ok Park-Cho, residing at 4655 Oakleigh Manor Drive, Powder Springs, GA 30127 and 1-207 Dong, A Apt., Chang-Dong, Tobong-Gu, Seoul, Korea, respectively, have invented certain new and useful improvements in IMPROVED INTERFERON POLYMER CONJUGATES for which I (we) are about to execute an application for a Patent of the United States

X which is executed on November 7, 1997
X which is identified by ROBERTS & MERCANTI, LLP, Docket No. 213.1020CIP2
_ which is filed on _____, Serial No. _____
X I (we) hereby authorize and request my (our) attorney, Roberts & Mercanti, LLP of P.O. Box 484 Princeton, New Jersey 08542-0484 to insert here in parentheses (Application number _____, filed _____) the filing date and application number of said application when known.

and WHEREAS, Enzon, Inc., 20 Kingsbridge Road, Piscataway, NJ 08854-3998, ASSIGNEE is desirous of obtaining the entire right, title and interest in, to and under the said invention and the said application:

NOW, THEREFORE, in consideration of the sum of One Dollar ($1.00) to me (us) in hand paid, and other good and valuable consideration, the receipt of which is hereby acknowledged, I (we), the said ASSIGNOR(S), have sold, assigned, transferred and set over, and by these presents do hereby sell, assign, transfer and set over, unto the said ASSIGNEE, its successors, legal representatives and assigns, the entire right, title and interest in, to and under the said invention, and the said United States application and all divisions, renewals and continuations thereof, and all Patents of the United States which may be granted thereon and all reissues and extensions thereof; and all applications for industrial property protection, including, without limitation, all applications for patents, utility models, and designs which may hereafter be filed for said invention in any country or countries foreign to the United States, together with the right to file such applications and the right to claim for the same the priority rights derived from said United States application under the Patent Laws of the United States, the International Convention for the Protection of Industrial Property, or any other international agreement or the domestic laws of the country in which any such application is filed, as may be applicable; and all forms of industrial property protection, including, without limitation, patents, utility models, inventors' certificates and designs which may be granted for said invention in any country or countries foreign to the United States and all extensions, renewals and reissues thereof;

AND I (WE) HEREBY authorize and request the Commissioner of Patents and Trademarks of the United States, and any Official of any country or countries foreign to the United States, whose duty it is to issue patents or other evidence or forms of industrial property protection on applications as aforesaid, to issue the same to the said ASSIGNEE, its successors, legal representatives and assigns, in accordance with the terms of this instrument.

AND I (WE) HEREBY covenant and agree that I (we) have full right to convey the entire interest herein assigned, and that I (we) have not executed, and will not execute, any agreement in conflict herewith.

AND I (WE) HEREBY further covenant and agree that I (we) will communicate to the said ASSIGNEE, its successors, legal representatives and assigns, any facts known to me (us) respecting said invention, and testify in any legal proceeding, sign all lawful papers, execute all divisional, continuing, reissue and foreign applications, make all rightful oaths, and generally do everything possible to aid the said ASSIGNEE, its successors, legal representatives and assigns, to obtain and enforce proper protection for said invention in all countries.

IN TESTIMONY WHEREOF, I (we) hereunto set my (our) hand and seal the day and year set opposite my (our) signature(s).

Date x Nov 7, 1997

x Carl W. Gilbert
(Signature of Inventor)

Carl W. Gilbert
(Typed Name of Inventor)

Date _____, 1997

_____
(Signature of Co-inventor)

Myung-ok Park-Cho
(Typed Name of Co-inventor)

A:\assignment.wpd

EXHIBIT X

Chronology of Regulatory Activities undertaken by Schering to support PEG-INTRON (Peginterferon alfa-2b) Powder for Injection under BB-IND #7173.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
06/02/97	BB-IND Application	BB-IND application. CD included pre-clin reports.
06/05/97	Letter from FDA	Letter from FDA acknowledging the receipt of our BB-IND for PEG IFN – Laudicina.
06/06/97	Letter to FDA	Pre BB-IND meeting minutes - Laudicina
06/18/97	Memo to Penelope Giles from Ms. J. Gress (FDA)	Clarification of timing of biopsy in BB-IND – Giles.
06/19/97	Memo from to Penelope Giles from Ms. J. Gress (FDA)	Request for availability PK and safety data to support the 1.5 µg /kg dose - Giles
06/23/97	Letter to FDA	Information sent to FDA (PK and safety) for 1.5 µg/kg proposed dose – Laudicina.
06/24/97	Letter to FDA.	Corrected fax sent to FDA with PK data for 1.5 µg/kg proposed dose in BB-IND 7173 – Laudicina.
06/26/97	Information Amendment: Chem/Microbiology	Provided additional stability data on PEG-IFN drug product (telephone request from CBER) – Feldman.
07/01/97	Memo from to Dr. P. Giles from Janet Gress (FDA)	Request for additional data from the MD PK study prior to initiating pivotal clinical study – Giles.
07/01/97	Memo from to Dr. P. Giles (FDA)from Ms. Janet Gress	Request for additional data from the MD PK study prior to initiating pivotal clinical study – Giles.
07/02/97	Memo to Ms. Janet Gress (FDA) from Dr. P. Giles	Confirm teleconference and attendees for 7/3/97 – Giles.
07/03/97	Memo to Vicky Tyson-Medlock (FDA) from M. Laudicina	FDA requested a teleconference to discuss safety issues regarding BB-IND 7173 –Laudicina.
07/03/97	Information amendment: Clinical	Response to FDA request for information – additional data was requested before it is safe to proceed with clinical trial – Laudicina.
07/10/97	Memo to Janet Gress/Dr. Anne Pilaro (FDA) from Dr. P. Giles	Follow-up on status of review for pivotal studies to proceed – Giles.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
07/10/97	Memo to Ms. Vicky Tyson-Medlock (FDA) from Ms. Laudicina	Receipt of additional information for BB-IND 7173 – Laudicina.
07/10/97	Memo to Ms. Janet Gress/Dr. Pilaro (FDA) from P. Giles	Follow-up on status of review for pivotal studies to proceed – Giles.
07/11/97	Memo to P. Giles from Janet Gress	Follow-up on status of review for pivotal studies to proceed.
07/15/97	Memo to Vicky Tyson-Medlock (FDA) from Malverna Laudicina	Schedule teleconference to discuss BB-IND 7173.
07/18/97	Memo to Ms. Vicky Tyson-Medlock (FDA) from Ms. Laudicina	Minutes of teleconference held on 7/18/97 to discuss BB-IND 7173 – Laudicina.
07/18/97	Memo to Ms. Vicky Tyson-Medlock (FDA) from M. Laudicina	Minutes of teleconference held to discuss BB-IND 7173 – Laudicina.
07/18/97	Letter to FDA Type: Technical	FMR Package – certification of analysis, HPSEC, chromatograms and SDS-PAGE gels for batches. (Feldman)
07/18/97	Letter to FDA	Protocol amendment and two new protocols
07/22/97	Memo to FDA from Ms. Laudicina	Scheduled teleconferences to discuss BB-IND's 7194 & 7173
07/23/97	Letter from FDA	Proposal for study design as requested by FDA on teleconference of 7/18/97 for Protocol 197-010. Laudicina
07/23/97	Memo to Vicky Tyson-Medlock (FDA) from Ms. Laudicina	Schedule teleconference to discuss study design. (Laudicina)
07/23/97	Memo to Janet Gress (FDA) from Dr. Giles	Scheduled teleconference to discuss study design. (Giles)
07/23/97	Letter from FDA	Letter for clinical hold for PEG-INTRON BB-IND in Hepatitis – Laudicina.
07/28/97	Memo to Ms. Vicky Tyson-Medlock (FDA) from Ms. Laudicina	Schedule teleconference to discuss study design – Laudicina.
07/28/97	Letter to FDA	Inquiries regarding clinical hold for BB-IND 7173 – Laudicina.
07/29/97	Teleconference with FDA	FDA lifts clinical hold on BB-IND 7173

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
07/29/97	Memo to Ms. Vicky-Medlock (FDA) from Ms. Laudicina	FDA requested a teleconference to discuss statistical issues regarding BB-IND 7173 – Laudicina.
07/29/97	Letter to FDA	Letter confirming clinical hold lifted and indicating we need to do 2 studies for registration per FDA request – Laudicina.
07/30/97	Memo to Ms. Janet Gress (FDA) from Dr. Giles	Confirmation by FDA of the receipt of our letter acknowledging the requirement for a second trial for PEG registration in the U.S. – Giles.
07/31/97	Memo to Ms. Vicky Tyson-Medlock (FDA) from Ms. Laudicina	Follow-up on amendment for CML protocol and CMC questions in the hold letter for Hepatitis – Laudicina.
08/01/97	Memo to Ms. Vicky Tyson-Medlock (FDA) from Ms. Laudicina	Effective dates for BB-IND 7173 and BB-IND 7194– Laudicina..
08/06/97	Letter to FDA	Copy of CRF for study C97-010 sent to FDA per Mr. Gross' request –Laudicina.
09/03/97	Letter to FDA	Document Types: Information Clinical Amendment: Protocol Amendments: New Protocols.
09/11/97	Protocol Amendment + New Investigator/Letter to FDA	Submission of investigator information for 2 phase 1 studies (renal insufficiency any young vs. old PK studies) – Giles.
09/22/97	Letter to FDA/ Adverse Drug Reaction (ADR)	Fallot ADR report type: 15 day BB-IND safety report.
10/18/98	Protocol Amendment	Submission of 12 U.S. & 3 int'l investigators for C97-010 & 197-010 respectively – Giles..
10/09/97	Information Amendment: Clinical	Transfer of sponsor obligations to Statprobe – Giles.
10/27/97	Letter to FDA Protocol Amendment/New Investigators	New investigators: Morgan, Hoefs, Carreno, Barcen-Marugan, Rodrigo-Saez – Giles
11/03/97	Letter to FDA re Clinical	FMR submission for Intron lyophilized batch 6-IFD-003 (Certificate of Analysis; test specifications; SDS-PAGE gel ID test) – Feldman.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
11/13/97	Letter from FDA	Official written notification of resolution of clinical hold. Note that the clinical hold was lifted on July 29, 1997 via telephone contact and confirmed via fax from SPRI to CBER the same day. (Giles).
11/19/97	Information Amendment/ Letter from FDA	Responses to comments in clinical hold letter dated July 23, 1997. (Comments 3-11). Responses include 1) updated drug substance characterization reports, 2) drug substance stability data and 3) finished product stability data – Feldman.
11/25/97	Letter to FDA	ADR Report - Adverse Drug Reaction -
12/04/97	Information Amendment: Chem/Microbiology	FMR document package (specifications: C of A; SDS-Page photo) for Intron batch (3 MIU vial) – Feldman.
12/09/97	Letter to FDA	Adverse Drug Reaction.
12/12/97	Letter to FDA	Protocol Amendment/New Investigator: Paul Martin, M.D. – Urquhart.
12/16/97	Letter to FDA	Adverse Drug Reaction
02/09/98	Letter to FDA	Adverse Drug Reaction
02/12/98	Memo to D. Urquhart from V. Tyson-Medlock (FDA)	Clinical
02/12/98	Memo to D. Urquhart from V. Tyson-Medlock (FDA)	Submission of Peg-Riba protocol to the BB-IND 7194.
02/12/98	Letter to FDA	Adverse Drug Reaction.
02/12/98	Letter to FDA	Adverse Drug Reaction
02/13/98	Memo to D. Urquhart from V. Tyson-Medlock (FDA)	Request from FDA to open a new BB-IND for Peg-Riba.
02/16/98	Letter to FDA	Adverse Drug Reaction
02/19/98	Letter to FDA	Adverse Drug Reaction
02/24/98	Letter to FDA	Adverse Drug Reaction
02/27/98	Letter to FDA	Advise FDA of actions regarding two severe cases of thrombocytopenia. New clinical guidelines to monitor patients implemented and protocol will be amended. Contains 11/11/98 minutes of DRAB meeting. Urquhart.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
03/10/98	Letter to FDA	Adverse Drug Reaction
03/12/98	Letter to FDA	Adverse Drug Reaction
3/13/98	Information Amendment: Chem/Microbiology	FMR package for PEG INTRON batch (Certificate of Analysis, HPSEC Chromatograms, SDS-Page Gel, Photos, Finished Product Release Specifications). Certificate of Analysis/Test P. Feldman.
03/19/98	Letter to FDA	Adverse Drug Reaction.
03/23/98	Letter to FDA	Request minutes of 1/25/95, 8/1/95 and 5/13/97 pre-BB-IND meetings with FDA. Urquhart.
03/24/98	Letter to FDA	Adverse Drug Reaction.
03/26/98	Letter to FDA	Adverse Drug Reaction
04/06/98	Letter to FDA	Adverse Drug Reaction.
04/10/98	Letter to FDA	Information Amendment: contains DRAB meeting (3/4/98) minutes and revised 1572 for Protocol Amendment: C97-010-04. Urquhart.
04/17/98	Letter to FDA	Adverse Drug Reaction
04/21/98	Letter from FDA	Contains copies of minutes from pre-BB-IND meeting of May 13, 1997 and August 1, 1995 meeting with FDA – Urquhart. (See our 3/23/98 letter.)
04/28/98	Letter to FDA	Adverse Drug Reaction.
05/01/98	Letter to FDA	Adverse Drug Reaction.
05/06/98	Letter to FDA	Adverse Drug Reaction.
05/07/98	Letter to FDA	Adverse Drug Reaction.
05/13/98	Information Amendment: Chem/Microbiology	FMR package for PEG-IFN batch (Certificate of Analysis, Specs, and HPSEC Chromatograms and SDS Page Photos) – Feldman
05/20/98	Information Amendment: Chem/Microbiology	New Strength 300mg vial.
05/26/98	Letter to FDA	Adverse Drug Reaction
05/27/98	Information Amendment: Clinical	DRAB meeting (April 18, 1998) minutes.
05/28/98	Letter to FDA	Adverse Drug Reaction.
06/02/98	Letter to FDA	Adverse Drug Reaction
06/02/98	Letter to FDA	Adverse Drug Reaction.
06/04/98	Letter to FDA	Adverse Drug Reaction.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
06/05/98	Letter to FDA	Adverse Drug Reaction
06/09/98	Letter to FDA	Adverse Drug Reaction.
06/10/98	Letter to FDA	Adverse Drug Reaction.
06/16/98	Letter to FDA	Adverse Drug Reaction.
06/19/98	Letter to FDA	Adverse Drug Reaction.
06/19/98	Letter to FDA	Adverse Drug Reaction.
06/25/98	Memo to Ms. Giles from J. Gress (FDA)	Follow-up on Hepatitis B Pediatric approval, PEG program and induction study – Giles.
06/25/98	Memo to P. Giles from W. Schwieterman/R. Lizambri (FDA)	Comment on induction protocol (C/198-169) amendment for genotypes 2 and 3.
06/25/98	Letter to FDA	Adverse Drug Reaction
06/26/98	Letter to FDA	Minutes of the 7/1/97 and 7/18/97 teleconferences – Urquhart.
06/26/98	Memo to Ms. Giles from J. Gress (FDA)	Follow-up on Hepatitis B approval, PEG program and induction study.
06/26/98	Information Amendment: Clinical	Minutes of June 1998 DRAB – Urquhart.
06/30/98	Memo to J. Gress (FDA) from P. Giles	Follow-up on Hepatitis B. Pediatric approval and teleconference on induction and PEG programs.
07/02/98	Letter to FDA	Adverse Drug Reaction.
07/07/98	Letter to FDA	Adverse Drug Reaction.
07/16/98	Letter to FDA	Adverse Drug Reaction.
07/23/98	Letter to FDA	Adverse Drug Reaction.
08/04/98	Letter to FDA	Protocol Amendments: Change in Protocol and Two New Investigators.
08/07/98	Letter to FDA	Protocol Amendments and New Investigator.
08/13/98	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report.
08/13/98	Letter to FDA	Request for minutes of teleconferences with FDA of July 18, & July 29, 1997.
08/20/98	Letter from FDA	Minutes of teleconferences with FDA of July 18, & July 29, 1997.
08/21/98	Information Amendment: Chem/Microbiology	FMR package including specifications and Certificate of Analysis, HPLC Chromatograms and SDS-Page Photos for Batch. Lawhon

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
08/21/98	Letter to FDA	Adverse Drug Reaction.
08/28/98	Letter to FDA	Adverse Drug Reaction.
09/15/98	Adverse Drug Reaction/ Letter to FDA	15 Day BB-IND Safety Report: Initial report for BB-IND 7173/ SCH 54031
09/17/98	Information Amendment: Chem/Microbiology	600 mcg vial and placebo formulation – Feldman.
09/18/98	Adverse Drug Reaction/ Letter to FDA	15 Day BB-IND Safety Report: Initial for Sch 54031 BB-IND 7173 – ADR Report Type: 15 Day BB-IND Safety Report.
09/21/98	Adverse Drug Reaction/ Letter to FDA	15 Day BB-IND Safety Report: Initial for BB-IND 7173 - Sch 54031 – ADR Report Type: 15 Day BB-IND Safety Report.
09/24/98	Adverse Drug Reaction/ Letter to FDA	15 Day BB-IND Safety Report – Follow-up – Fallot ADR Report Type: 15 Day BB-IND Safety Report.
09/28/98	BB-IND Annual Report/ Submitted for period: 07/31/97 – 07/30/98	BB-IND Annual Report for Sch 54031, BB-IND 7173 – Urquhart.
09/30/98	Adverse Drug Reaction/ Letter to FDA	Mannino ADR Report Type: 15 Day BB-IND Safety Report.
09/30/98	Letter to FDA	Response to request for information – Statistical Analysis Plan for C/197-010
10/02/98	Letter to FDA	Information Amendment: Clinical.
10/08/98	Memo to V. Tyson-Medlock (FDA) from D. Urquhart	Teleconference to discuss the Statistical Analysis Plan for the PEG study C/197-010.
10/20/98	Memo to V. Tyson-Medlock (FDA) from D. Urquhart	Follow-up on 9/30/98 letter to FDA containing an explanation of Statistical Analysis Plan for ongoing study requested by Dr. Schweiterman (FDA) – Urquhart.
10/26/98	Memo to V. Tyson-Medlock/D. Urquhart from D. Urquhart/J. Gress (FDA)	PEG-INTRON – FDA contact 10/27/98
11/03/98	Adverse Drug Reaction/ Letter to FDA	Cobert ADR Report Type: 15 Day BB-IND Safety Report.
11/03/98	Adverse Drug Reaction/ Letter to FDA	Cobert ADR Report Type: 15 Day BB-IND Safety Report.
11/05/98	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
11/06/98	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
11/06/98	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
11/11/98	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
11/13/98	Information Amendment: Chem/Microbiology	FMR Package for PEG-IFN batch and Intron Lyo Batch
12/02/98	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
12/07/98	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
12/08/98	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
01/05/99	Memo to D. Urquhart from J. Tiwari (FDA)	Follow-up on request for data to confirm modeling in in-treatment virology endpoints.
01/20/99	Memo to D. Urquhart from J. Tiwari (FDA)	Follow-up requests for datasets to support modeling of 24 week virology.
01/27/99	Memo to V. Tyson-Medlock (FDA) from J. Aoyagi	Pre-BLA meeting request.
02/22/99	Information Amendment: Chem/Microbiology	FMR Package for PEG-IFN batches and Intron solution for injection batch
02/23/99	Letter to FDA	Meeting request for end of Phase III/Pre-BLA.
02/25/99	Memo to V. Tyson-Medlock (FDA) from J. Aoyagi	PEG Monotherapy Pre-BLA meeting request.
03/03/99	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
03/04/99	Letter to FDA	Pre-BLA meeting request for PEG Monotherapy; 24 week in-treatment report (C/I97-010) and 48 week safety summary (C/I97-010) enclosed.
03/10/99	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
03/11/99	Memo to V. Tyson-Medlock (FDA) from J. Aoyagi	Follow-up on meeting request (BB-IND #7173) and on response concerning PCR measurements (BB-IND #7572)

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
03/12/99	Memo to D. Urquhart from J. Gress	J. Gress memo to Schweiterman/Weiss of FDA that SPRI sent the modeling data requested by Tiwari to validate the in-treatment endpoint (sent to BB-IND 7572 PEG/Riba).
03/12/99	Memo to J. Gress/V. Tyson-Medlock (FDA) from D. Urquhart	FDA contact on PEG/RIBA – BB-IND 7572 and PEG Intron – BB-IND 7173.
03/12/99	Memo to Vicky Tyson-Medlock (FDA) from Penny Giles	Follow-up on request for pre-BLA meeting and on PEG/RIBA responses Re: 2 PCR measurements
03/22/99	Information Amendment: Chem/Microbiology	FMR package for Intron HSA free solution batch
03/24/99	Memo to J. Aoyagi from V. Tyson-Medlock of FDA	End of Phase III conference call.
04/15/99	Memo to Vicky Tyson-Medlock (FDA) from Giles	Follow-up on scheduling.
04/16/99	Memo to Vicky Tyson-Medlock (FDA) from Giles	Follow-up on scheduling 1) a meeting on the Intron A label; 2) a teleconference on ECOG data; 3) Hepatitis database and modeling.
04/16/99	Memo to Vicky Tyson-Medlock (FDA) from Giles	To follow-up on arranging a meeting (how the Intron A label could be improved) and two teleconferences (to discuss FDA's request to include ECOG data for the Melanoma indication; to discuss HCV modeling/in-treatment endpoint).
04/21/99	Information Amendment Chem/Microbiology	FMR package for PEG-IFN batches 8-IQA-101 and 8-IQC-101
04/22/99	Memo to W. Schweiterman (FDA) from P. Giles	BLA for PEG-Intron.
04/23/99	Memo to W. Schweiterman (FDA) from P. Giles	PEG-Intron
04/23/99	Memo to Vicky Tyson-Medlock (FDA) from Giles	Follow-up on scheduling.
04/26/99	Memo to Vicky Tyson-Medlock (FDA) from D. Urquhart	SPRI attendees at 4/22/99 teleconference with FDA.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
05/04/99	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
05/06/99	Memo to D. Urquhart from A. Pilaro (FDA)	Design issues – PEG.
05/06/99	Information Amendment: Chem/Microbiology	Drug Product Attachments: Stability, Drug Product specifications, Drug Product Analytical Procedures, Drug Substance Attachments: Validation Summaries/Protocols/Drug Substance Specification, Drug Substance Analytical Proc
05/06/99	Information Amendment: Chem/Microbiology	FMR package for HAS free solution batch 8-IO/b-061 Drug Product Attachments: Drug Product Specifications, Certificate of Analysis/Test P
05/07/99	Memo to W. Schwieterman (FDA) from P. Giles	Summary of 5/7/99 Teleconference; Purpose of call was to alert Dr. Schwieterman to the early IL-10 efficacy data in chronic hepatitis C. Also, obtain additional feedback regarding FDA;s dissatisfaction with the Intron A label. Also, inquire if they had further thoughts regarding our database and modeling for the early week 24 during treatment endpoint.
05/10/99	Memo to Richard Morrissey (FDA) from Rachael Steiner	Summary of 5/10/99 Teleconference regarding the PEG repro monkey study commitment and study design.
05/12/99	Letter from FDA	FDA minutes from April 22, 1999 teleconference.
05/13/99	Protocol Amendment	Info. Amendments
05/18/99	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
05/27/99	Memo to V. Tyson-Medlock (FDA) from P. Giles/Steiner/Lamendola	FDA/Schering working meeting to discuss the Intron A PI and ways to make it more user friendly.
05/27/99	Memo to FDA from Steiner	FDA/Schering working meeting to discuss the Intron A PI and ways to make it more user friendly.
05/27/99	Letter to FDA	Information Amendment, Protocol Amendment and New Investigator.
05/28/99	Letter to FDA	Confirmation of receipt of serial no. 098 previously submitted on 5/6/99
06/01/99	Memo to A. Pilaro (FDA) from D. Urquhart	Fax to tox reviewer to obtain comments on design of (monkey) protocol (estrus effects and hormone levels).

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
06/08/99	Memo to D. Urquhart from V. Tyson-Medlock	Discussion of pre-BLA meeting with FDA, CSO stated that office policy requires the submission of unblinded data for this.
06/08/99	Letter to FDA	Copy of NGI letter submitted to FDA to allow FDA to cross reference their applications (PLA) in support of BB-IND 7173 and 7572. Letter was also submitted to BB-IND 7572 on 6/8/99.
06/08/99	Letter to FDA	Letter to request teleconference with Michael Fauntleroy of FDA who is in charge of electronic submissions at CBER.
06/15/99	Information Amendment	FMR Package/Drug Product Attachments:/Drug Product Specifications
06/16/99	Protocol Amendment: New Investigator	Revision of 1572's and site personnel, also informed FDA that Rachael Steiner is new contact person.
06/18/99	Memo to Rachael Steiner from M. Fauntlerroy (FDA)	Cancellation of teleconference to discuss PEG BLA electronic submission.
06/22/99	Memo to Rachael Steiner from M. Fauntlerroy (FDA)	Preferred dates for teleconference on electronic PEG BLA, draft guidance on electronic submissions due out 7/99.
06/25/99	Memo to Rachael Steiner from M. Fauntlerroy (FDA)	Confirmation of July 9, 1999 3 P.M. teleconference.
06/28/99	Information Amendment: Clinical/Letter to FDA	Correction of site information – deletion of subinvestigator.
06/29/99	Letter to FDA	Inform FDA of administrative clinical holds on site C97-010-38 (Dr. Hoefs) by site's IRB.
07/01/99	Memo to D. Urquhart from A. Pilaro (FDA)	Verbal agreement from FDA that PEG will not receive a RTF for not containing the estrus cycle/hormone monkey study requested by FDA.
07/01/99	Memo to Anne Pilaro (FDA) from R. Steiner	Requested update on FDA's providing comments on 6/1/99 fax (protocol concept for monkey study).
07/01/99	Letter to FDA	Hardcopy of 6/1/99 fax to Anne Pilaro sent – protocol concept for female cyno monkey study.
07/02/99	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
07/08/99	Letter to FDA	Cross referencing BB-IND 7572, Serial 023 which contains updated SOPS for NGI's PCR assay.
07/09/99	Memo to R. Steiner/P. Giles from M. Fauntleroy (FDA)	Summary of data submission requirements provided via e-mail to Schering from M. Fauntleroy (FDA).
07/09/99	Memo to M. Fauntleroy (FDA) from R. Steiner	E-mail containing list of Schering attendees for the 7/9/99 electronic BLA submission teleconference.
07/09/99	Memo to M. Fauntleroy (FDA) from R. Steiner	Summary of 7/9/99 teleconference on electronic issues for PEG BLA submission.
07/14/99	Memo to Destry Sillivan/Tyson-Medlock (FDA) from Rachael Steiner	Messages for follow-up on pregnancy registry response and to request feedback on PEG monkey fertility study.
07/16/99	Information Amendment: Pharm/Tox/Letter to FDA	Pre-clin reports
07/19/99	Letter to FDA	CALA letter of understanding.
07/21/99	Memo to Rachael Steiner from Vicky Tyson-Medlock (FDA)	V. Tyson-Medlock did not locate any comments from Dr. Pilaro regarding the PEG monkey study.
07/21/99	Memo to Giles/Steiner from Fleischer/Sillivan (FDA)	Low does not discussed within agency, pregnancy response looks goods, send letter of understanding for pediatric agreement, food effect no commitment letter on the way, PEG submissions and antiviral activity discussed.
07/21/99	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
07/23/99	Adverse Drug Reaction/ Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
07/26/99	Letter to FDA	Request for revisions to monkey study protocol concept (copy also faxed to Anne Pilaro).
07/27/99	Memo to Dr. Schwieterman (FDA) from Penny Giles	PEG in HIV, PEG BLA feedback on logistics for the submission. No review yet of early Intron AE section draft.
07/27/99	Memo to Dr. Schwieterman (FDA) from Penny Giles	We requested an end of Phase 1 meeting to discuss the use of PEG Intron A in HIV. We feel we need a meeting since we now have Phase 1/2 results and wish to discuss a proposed pivotal program to register this for HIV treatment.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
07/27/99	Letter to FDA	Response to CBER request to provide replacement pages i.e. corrected reference STD. Chromatograms included in June 15, 1999 amendment (Serial No. 104)
08/02/99	Letter to FDA	Letter of understanding based on 4/22/99 telcon – tox, clinical, BLA content issues addressed.
08/03/99	Memo to R. Steiner from M. Fauntleroy (FDA)	Statistical hardcopy is necessary for the archival copy even though it duplicates the clinical.
08/03/99	Adverse Drug Reaction Letter to FDA	ADR Report Type: 15 Day BB-IND Safety Report
08/04/99	Memo to A. Pilaro (FDA) from R. Steiner	CYNO monkey study teleconference set up for August 6, 1999.
08/05/99	Memo to Anne Pilaro (FDA) from Rachael Steiner	Fax to Anne Pilaro containing list of attendees for the August 6, 1999 10:30 A.M. teleconference on the CYNO monkey study.
08/06/99	Memo to Pilaro/Serabian (FDA) from Schering	Finalize study design for tox Cyno monkey study; access PEG's effects on hormone/estrus cycles.
08/10/99	Information Amendment: Clinical/Letter to FDA	Protocol Amendment: New Investigator Updated IB, new principal investigator, site correction for 09.
08/24/99	Letter to FDA	Letter of cross reference to BB-IND 7572, Serial No. 033 submission of NGI PCR assay information.
08/25/99	Memo to V. Tyson-Medlock (FDA) from R. Steiner	Requested information on August 2, 1999 submission – no ISS/ISE, one large pivotal trial.
08/31/99	Information Amendment: Chem/Microbiology	Regulatory guidance meeting request plus copies (13) at CMC briefing document.
09/01/99	Memo to M. Fauntlerroy (FDA) from R. Steiner	Discussion with M. Fauntleroy on procedure for setting up a technical meeting and cala demo issues.
09/01/99	Memo to M. Fauntlerroy (FDA) from Rachael Steiner	Discussion with M. Fauntleroy on procedure for setting up a technical meeting and cala demo issues.
09/01/99	Letter to FDA	Request for pediatric studies deferral.
09/09/99	Letter to FDA	Request for review of proprietary name: PEG-Intron
09/13/99	Information Amendment: Clinical	Information Amendments/Protocol Amendments
09/14/99	Letter to FDA	Request an agenda for pre-BLA meeting.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
09/17/99	Memo to R. Steiner from V. Tyson-Medlock (FDA)	Schering needs to provide 72 week primary endpoint data tables in the briefing book in order to have the meeting.
09/17/99	Memo to R. Steiner from V. Tyson-Medlock (FDA)	Discussion on what 72 week data would be provided in briefing book – Schering stated key efficacy and safety tables.
09/20/99	Memo to Rachael Steiner from V. Tyson-Medlock (FDA)	Scheduling of pre-BLA meeting for 11/2/99 from 12:30 – 2:00 P.M. and request for update on request for pediatric deferral.
09/21/99	BB-IND Annual Report to FDA	Ref. Serial No. 070 and 097 for CMC Changes. Submitted for period 7/31/98 to 7/30/99.
09/24/99	Memo To: P. Giles From: W. Schwieterman (FDA)	Clarification of Request for 2 Page Summary of Pivotal PEG Hepatitis Trial and Follow Up on the Response for Intron A Phase IV Commitments.
09/24/99	Letter from FDA	Comments on Monkey Repro Study.
09/27/99	Memo To: W. Schwieterman (FDA) From: Penny Giles	Primary Efficacy Endpoint Data Provided as Requested Via Fax.
09/27/99	Letter from FDA	Meeting Announcement for Pre-BLA – 11/2/99, 12:30 – 2 P.M. – Includes List of Attendees.
09/28/99	Letter to FDA	Letter of Cross Reference – Dr. Afdahl.
09/29/99	Memo To: Rachael Steiner From: V. Tyson-Medlock (FDA)	Confirmation of November 2, 1999 Meeting from 12:30 – 2:00 P.M. and 20 Copies of Pre-BLA Briefing Book.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.
10/01/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR Report Type: 15 Day BB-IND Safety Report.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
10/04/99	Letter to FDA	CALA Demo CD Provided to Agency for PEG Electronic Submission.
10/07/99	Memo To: Janet Gress (FDA) From: Rachael Steiner	Fax to Janet Gress – PEG Primary Endpoint Efficacy Tablets.
10/13/99	Memo To: R. Steiner From: V. Tyson-Medlock (FDA)	Briefing Books have been distributed as well as the Electronic Submission Demo.
10/15/99	Memo To: R. Steiner From: S. Sickafuse (FDA)	Request for Information on Intron Label Review.
10/22/99	Letter to FDA	Monkey Study Design Response to 9/24/99 Letter from FDA.
10/25/99	Memo To: Dr. Earl Dye From: Dr. Diane Zezza (FDA)	Pre-BLA Meeting Minutes on CMC Issue (10/20/99).
10/27/99	Memo To: Rachael Steiner From: M. Fauntleroy (FDA)	M. Fauntleroy requested teleconference with Schering to Discuss Electronic Demo Provided; Scheduled for 11/1/99.
10/28/99	Memo To: R. Steiner From: V. Tyson-Medlock (FDA)	Small Revision to Label Incorporating Rebetron References Requested; Indent Pregnancy Category X Statement and Revise Slightly: No Further Information Needed for Pre-BLA Meeting.
10/29/99	Memo To: R. Steiner From: S. Sickafuse (FDA)	Update on Intron Label Review Revised as Per the Agency's Request.
11/01/99	Memo To: M. Fauntleroy (FDA) From: R. Steiner	List of Teleconference Attendees for 11/1/99 Electronic Submission Teleconference.
11/01/99	Memo To: Rachael Steiner From: Michael Fauntleroy (FDA)	Teleconference to Dismiss Issues with PEG Electronic Submission Demo Provided to Agency.
11/02/99	Memo To: Dr. Joe Bekisz (FDA) From: Stuart Feldman	Follow-up to Pre-BLA Teleconference Re: Endotoxin Levels in PEG-IFN Drug Substance.
11/02/99	Memo	Pre-BLA Meeting Minutes for PEG-Intron Monotherapy.
11/05/99	Memo To: FDA Document Office From: Rachael Steiner	User Fee Assigned for PEG-BLA.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
11/05/99	Memo To: V. Tyson-Medlock (FDA) From: R. Steiner	Items Needed for BLA Submission and Confirmation on Stats Files Requested (Send to BB-IND and Desk Copy for Tiwari).
11/09/99	Memo To: Rachael Steiner From: A. Pilaro (FDA)	Teleconference on cyno monkey study no longer needed, Schering's 10/22/99 response to FDA stating only hematology to be done is correct.
11/09/99	Letter to FDA	Statistical report, data and programs requested at pre-BLA meeting; desk copy to Dr. Tiwari.
11/17/99	Letter to FDA	Demo case report form and scanned document. Schering's Nov. 1, 1999 teleconference meeting minutes.
11/18/99	Information Amendment: Chem/Microbiology	FMR package – 600 μg vial, drug product attachments, drug batch numbers, information amendments/protocol amendments.
11/19/99	Memo To: Rachael Steiner From: William Schwieterman/FDA	Teleconference meeting minutes.
11/22/99	Information Amendment: Clinical	Updated 1572 and removal of subinvestigator for site C97-010-15.
11/24/99	Letter to FDA	Sample Electronic SAS transport files and blank CRF; desk copy to M. Fauntleroy.
11/29/99	Memo To: Rachael Steiner From: M. Fauntleroy (FDA)	BLA submission of electronic information.
11/29/99	Memo To: Ms. Steiner From: Ms. Tyson-Medlock (FDA)	BLA submission logistics.
11/29/99	Letter to FDA	Schering's meeting minutes from November 2, 1999 pre-BLA meeting and statistical history submitted.
11/30/99	Memo To: Penny Giles From: J. Frey (FDA)	Electronic BLA submission – no restrictions on timing.
12/01/99	Letter to FDA	Statistical summary document for Intron monotherapy, REBETRON and PEG; BB-IND and desk copies to Tiwari, Schwieterman and Gress.
12/02/99	Letter from FDA	FDA official meeting minutes from the November 2, 1999 pre-BLA meeting.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
12/03/99	Memo To: W. Schwieterman (FDA) From: P. Giles	Statistical modeling with week 24 virologic endpoint.
12/03/99	Memo To: W. Schwieterman (FDA) From: P. Giles	Statistical modeling with week 24 virologic endpoint.
12/06/99	Information Amendment: Clinical	Updated 1572's provided – info. amendments/ protocol amendments.
12/09/99	Letter to FDA	Cross reference letter to BB-IND 7572 monkey toxicology studies (serial no. 042) pre-clin reports: P-6671, P-6752.
12/13/99	Letter from FDA	CBER instructions for submission of large license and BB-IND applications.
12/13/99	Memo To: Rachael Steiner From: M. Fauntleroy (FDA)	BLA submission of electronic statistics information.
12/13/99	Memo To: Jules Meisler (FDA) From: Rachael Steiner	BLA submission logistics with document room receipt.
12/14/99	Memo To: P. Giles From: V. Tyson-Medlock (FDA)	Send copy of Intron phase IV commitment submission. Scheduling of the teleconference to discuss modeling. Submission of corrections to Pre-BLA minutes.
12/14/99	Memo To: P. Giles From: V. Tyson-Medlock (FDA)	Send copy of Intron phase IV commitment submission. Scheduling of the teleconference to discuss modeling. Submission of corrections to Pre-BLA minutes.
12/16/99	Letter to FDA	Fax with BLA TOC for agency's review and possible request for additional hard copies.
12/16/99	Memo To: V. Tyson-Medlock (FDA) From: Rachael Steiner	Let FDA know TOC faxed and to see if any additional hardcopies will be needed.
12/17/99	Letter to FDA	Fax to Dr. Tiwari with tables of combined endpt., PCR and ALT response at treatment week 24, end of treatment and at end of follow up.
12/17/99	Memo To: P. Giles From: Dr. Tiwari/ Dr. Schwieterman (FDA)	Discussion of stat model – PEG Intron.
12/20/99	Memo To: Jules Meisler (FDA) From: Rachael Steiner	BLA submission logistics with document.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
12/20/99	Memo To: R. Steiner From: B. Schwieterman (FDA)	Requested REBETRON information for response needed prior to teleconference on modeling acceptance.
12/20/99	Memo To: Rachael Steiner From: Anne Pilaro (FDA)	Request for P-6671 and P-6752 PEG/RIBA monkey tox study protocols.
12/21/99	Letter to FDA	Provided via fax tables containing the Rebetron efficacy data with sabgroups identified – high viral load, low viral load, cirrhosis, and genotype.
12/21/99	Memo To: Steiner/Heft/ Ling From: Dr. J. Tiwari (FDA)	Discussion on the statistical modeling predictions.
12/21/99	Memo To: Rachael Steiner From: Dr. J. Tiwari (FDA)	CBER statistician requested confirmation he had received all documentation and wanted to know why data published in New England Journal of Med Article was different than Modeling (Journal Article Patient Subset of Database).
12/22/99	Memo To: Vicky Tyson-Medlock (FDA) From: Rachael Steiner	Alert agency that the PEG BLA is on the way.
12/23/99	Letter to FDA	Request for revisions to FDA's official meeting minutes of November 2, 1999 pre-BLA meeting.
12/23/99	Adverse Drug Reaction/ Letter to FDA	Clinical ADR report type: 15 day BB-IND safety Report.
01/04/00	Memo To: Dr. Schwieterman (FDA) From: Dr. P. Giles	Discussion on the statistical modeling predictions. Scheduling teleconference.
01/04/00	Memo To: Dr. Schwieterman (FDA) From: Dr. Giles	Discussion on the statistical modeling predictions.
01/05/00	Memo To: Rachael Steiner From: V. Tyson-Medlock (FDA)	BLA number assignment, 99-1488.
01/06/00	Information Amendment: Clinical	New subinvestigator at site C97-010-15 info. amendments/protocol amendments.
01/07/00	Letter to FDA	IIS plans for statistical help and transfer of obligations to Lark.
01/21/00	Information Amendment: Chem/Microbiology/ Letter to FDA	FMR package (certificate of analysis ("C of A"); specs; chromatograms and gel photos).

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
01/24/00	Information Amendment: Pharm/Tox	Mouse micronucleus in PEG report, information amendments/protocol amendments.
02/10/00	Information Amendment: Chem/Microbiology	FMR package. (Drug product attachments C of A, Drug Batch Nos.)
02/17/00	Information Amendment: Clinical	Updated site information location and sub-investigator info. amendments/protocol amendments.
03/16/00	Information Amendment: Pharm/Tox/Letter to FDA	Amended tox report on PEG/pre-clin reports.
04/07/00	Letter to FDA	Cross reference letter for Ronald Bukowski, M.D. for study entitled "A Phase I Trial of PEG Intron And IL-2 In Patients With Metastatic Renal Cell Carcinoma".
04/12/00	Information Amendment: Chem/Microbiology	FMR document package for various PEG-Intron batches; info. amendments/protocol amendments.
04/18/00	Memo To: Rachael Steiner From: L. Marzella (FDA)	Request for pediatric information for REBETRON.
04/21/00	Information Amendment: Clinical	REBETRON pediatric information; interferon P00-018 CSR (week 12) information amendments/protocol amendments.
04/24/00	Memo To: W. Schwieterman/ M. Thornton (FDA) From: Penny Giles	Investigator initiated studies (GISH); European HRD for PEG and RIBA.
4/26/00	Letter to FDA	Cross reference letter for Dr. Zeev Estrov and Dr. Francis Giles.
05/01/00	Letter to FDA	Cross reference letter for Dr. Razelle Kurzrock.
05/02/00	Memo To: W. Schwieterman (FDA) From: P. Giles	PEG+RIBA data release, provide 24-week report to FDA, heads up on labeling amendment for PEG monotherapy, reported delay in revision to Intron A label.
05/02/00	Protocol Amendment: New Protocol.	Clinical info. amendments/protocol amendments – new protocol.
05/03/00	Letter to FDA	Cross reference letter for Gary Schiller, MD.
05/04/00	Letter to FDA	FMR document package for Intron drug batch numbers.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
05/16/00	Protocol Amendment: New Protocol/New Investigator	"Effects of multiple-dose PEG-Intron on the activity of drug metabolizing enzymes in healthy volunteers".
05/23/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
05/23/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
05/25/00	Information Amendment: Chem/Microbiology	FMR package drug product attachments, drug batch numbers.
05/25/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
05/25/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
05/26/00	Information Amendment: Chem/Microbiology	Changes to manufacturing processes for drug product including addition of a pre-filtration step (0.45 μm filter) prior to final sterile filtrations deletion of safety test in drug product specifications.
05/30/00	Letter to FDA	Fax list of teleconference attendees for the 5/30/00 Investigator Initiated Study ("IIS") discussion.
05/30/00	Memo To: Rachael Steiner From: Schwieterman (FDA)	Teleconference to discuss the PEG and PEG-RIBA IIS.
05/30/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
06/01/00	Information Amendment: Clinical Protocol Amendment: Change in Protocol	Representative labeling and protocol amendment #1, P01-504 (one volume).
06/09/00	Letter to FDA	Cross reference letter for Joseph Clark, MD.
06/12/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
06/12/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
06/13/00	Information Amendment: Chem/Microbiology	Revised HPSEC chromatograms as per CBER request – drug batch numbers.
06/16/00	Letter to FDA	Comparison of Roche PEGASYS to REBETRON for treatment of cirrhotics —priority review should not be grated.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
06/21/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
6/22/00	Information Amendment: Pharm/Tox	PEG repro study – two volumes – pre-clin reports.
06/23/00	Letter to FDA	Cross reference letter for Dr. Lewis.
06/28/00	Protocol Amendment: Change in Protocol	Information amendments/protocol amendments – change in protocol.
07/11/00	Information Amendment: Clinical	C/197-010 PPK report 0068050.
07/14/00	Information Amendment: Chem/Microbiology	FMR package for batch 0-IQB-101 – drug product attachments – drug batch numbers.
07/18/00	Memo To: R. Steiner From: Dr. Schwieterman (FDA)	PEG monotherapy pediatrics teleconference.
07/21/00	Letter to FDA	SAE at PN HOEF's site discovered after study completion.
08/15/00	Letter to FDA	Letter of cross reference for the division of AIDS/PATCG.
08/16/00	Letter to FDA	Data as desk copies for reviewers of the pediatric HIV trial under BB-IND 9197 (non-Schering), 018 week 12 report, CSR for I97-078.
08/23/00	Protocol Amendment: New Investigator	Information amendments/protocol amendments – 4 new investigators.
08/28/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
08/30/00	Letter to FDA	Fax to Anita O'Connor containing preclinical tox. information for PEG-Intron.
08/31/00	Memo To: Penny Giles From: Fiona Mingioli/ W. Schwieterman (FDA)	Scheduling teleconference for large IIS study for PEG-Intron plus Ribavirin in naives.
09/01/00	Memo To: Penny Giles From: Fiona Mingioli/ W. Schwieterman (FDA)	Scheduling teleconference for large IIS study for PEG-Intron plus Ribavirin in naives.
09/06/00	Memo To: Fiona Mingioli (FDA) From: Penny Giles	Scheduling teleconference for large IIS study for PEG-Intron plus Ribavirin in naives.
09/07/00	Memo To: Rachael Steiner From: Janet Gress (FDA)	FDA teleconference on the large IIS study – Jacobson and Brown also present.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
09/18/00	Memo To: Rachael Steiner From: V. Tyson-Medlock (FDA)	Agency follow-up on pediatric proposal for PEG monotherapy.
09/18/00	Memo To: R. Steiner From: L. Marzella (FDA)	Agency follow-up on PEG simplified dosing regression analysis, request for Intron information on hypotension and confirmation of the IL-10 teleconference.
09/18/00	BB-IND Annual Report 07/31/99-07/30/00	PEG Intron IB (8/00), study updates and preclinical data included.
09/26/00	Information Amendment: Chem/Microbiology	FMR package – drug batch numbers.
09/28/00	Information Amendment: Chem/Microbiology	FMR package – drug batch numbers.
09/29/00	Letter to FDA	Corrected table of contents page for BB-IND amendment, serial no. 206.
11/02/00	Protocol Amendment: New Investigator	Information amendments/protocol amendments – 3 new investigators.
11/08/00	Letter to FDA	Notify FDA of change of contact of information.
11/10/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
11/21/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
11/22/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
12/04/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
12/06/00	Adverse Drug Reaction/ Letter to FDA	ADR report type: 15 day BB-IND safety report.
12/12/00	Letter to FDA	Protocol amendment: new investigator.

EXHIBIT XI

Chronology of Regulatory Activities undertaken by Schering to support PEG-INTRON™ (Peginterferon alfa -2b) Powder for Injection BLA #99 1488

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
12/22/99	Letter to FDA	Original BLA Submission
12/22/99	Letter to FDA	User Fee Payment For PEG BLA
1/06/00	Letter from FDA	Assignment of BLA #99-1488
1/14/00	Memo to Stuart Feldman from Ms. Debbie Trout (FDA)	License Status of PEG-INTRON Manufacturing Facilities.
1/18/00	Memo to Stuart Feldman from Debbie Trout (FDA)	Request for Peg-Intron Production Schedule through July 2000.
1/20/00	Memo to Ms. Debbie Trout (FDA) from Dr. Stuart Feldman	Production Schedule for Peg-Intron Drug Substance and Drug Product through July 2000.
2/04/00	Letter to FDA	Pediatric Proposal-Waiver Peg Memo Sudies, Do PEG and RIBA Only.
2/07/00	Memo to V. Tyson-Medlock (FDA) from R. Steiner	Standard 10 month review given to BLA; no refusal to file issues.
2/09/00	Memo to R. Steiner from Tyson-Medlock/Pilaro (FDA)	Agency requested approximate time of delivery for the Peg Monkey Study Report-June target.
2/14/00	Letter to FDA	4-Month safety update content proposal.
2/16/00	Letter from FDA	Peg application is acceptable for filing.
2/18/00	Memo to Rachael Steiner from Tyson-Medlock (FDA)	Peg-Intron name approved, letter stating application is acceptable for filing is on the way
2/24/00	Memo to V. Tyson-Medlock (FDA) from Rachael Steiner	Requested update on review of the 4 month safety update.
3/10/00	Memo to Rachael Steiner from Vicky Tyson-Medlock (FDA)	4-Month Safety Update Proposal still pending. Agency plans to respond by 3/17/00 or O.K. to go as presented.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
3/13/00	Memo to Ms. Debbie Trout (FDA) from Stuart Feldman	Revised Production Schedule for Peg-Intron (Drug Substance/Drug Product)
3/17/00	Memo to Rachael Steiner from Vicki Tyson-Medlock (FDA)	Only Ms. Gress responded to Schering's Proposal for 4-month safety update. She stated O.K. and Schering should contact her if guidance on format is needed.
3/20/00	Memo to Rachael Steiner from Janet Gress (FDA)	Schering 2/14/00 proposal for what information to include in the 4-month safety update is acceptable.
3/29/00	Memo to P. Giles from Louis Marzella (FDA)	Peg Intron/Hepatitis
3/31/00	BLA Amendment to FDA	Labeling to replace 1 ML. Diluent Vial with 5 ML. Vial.
4/07/00	Memo to Vicky Tyson Medlock (FDA) from Rachael Steiner	Ms. Tyson-Medlock requested additional copies of the March 31, 2000 submission; no comments on the pediatric 2/4/00 submission.
4/07/00	Memo to Dr. Tiwari (FDA) from Giles/Steiner/Ling	Dr. Tiwari needs assistance in using the SAS programs and data sets provided as part of the BLA.
4/07/00	Memo to Dr. Louis Marzella (FDA) from Giles/Steiner	Summary of phone contact 4-month safety report requirements and future labeling amendment.
4/07/00	Memo to M. Fauntleroy (FDA) from P. Giles	Request from Schering for M. Fauntleroy to provide technical support to the statistical reviewer; Dr. Tiwari as he is having difficulties in loading/using the SAS datasets and programs.
4/10/00	Memo to Rachael Steiner from Dr. Tiwari (FDA)	Dr. Tiwari has requested assistance in helping load all 10 CD's of the BLA submission onto his desktop computer.
4/10/00	Letter to FDA	Fax to Dr. Marzella of FDA regarding telecon minutes and bulleted agreements; 4-month safety update, future.
4/11/00	Letter to FDA	Fax to Dr. Tiwari containing the SAS file/datasets and instructions for the BLA. CD10 cover copies as this one is needed to use the statistical files.
4/11/00	Memo to Rachael Steiner from Dr. Marzella (FDA)	Dr. Marzella requested information on the status of the pediatric programs for the use of PEG-INTRON therapy with and without ribavirin.
4/12/00	Memo to Rachel Steiner from M. Fauntleroy	Schering requested to know number ot CDS required for stats submission – need 3 CDS

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
4/12/00	Memo to P. Giles from M. Fauntleroy (FDA)	Provide Airbill and carrier name to M. Fauntleroy for the statistical CD submission
4/12/00	Memo to Rachel Steiner from Dr. Mazrella (FDA)	Dr. Marzella requested information on the status of the pediatric programs for the use of Peg-Intron with and without ribavirin
4/14/00	Memo to M. Fauntleroy (FDA) from Rachel Steiner	Schering has sent the replacement stats CDC (Airbill and carrier information provided)
4/14/00	Letter to FDA	Replacement CD 10 of 10 – Corrected Statistical Files
04/18/00	Memo to Rachel Steiner from L. Marzella	Request for pediatric information for Rebetron
04/20/00	Letter to FDA	4 Month safety update – Includes Hepatitis, HIV MS and Oncology (two volumes)
05/02/00	Memo to W. Schwieterman (FDA) from P. Giles	Peg & Riba data release provide 24-week report to FDA, Heads up on labeling amendment for Peg Monotherapy, reported delay in revision to Intron A label
05/02/00	Memo to Stuart Feldman from Debbie Trout	Pre-Approval inspection for Peg Intron
05/05/00	Memo to Rachel Steiner from Dr. Marzella (FDA)	Dr. Marzella requested the 24 week in treatment report, information on the carcinogenicity studies for Ribavirin and critical ribavirin study reports should be provided as part of the PEG/Riba BLA
05/15/00	Letter to FDA	Advanced data from monkey study (SN-99331) provided to FDA prior to final report (desk copy to Anne Pilaro).
05/23/00	Memo to Rachel Steiner from Dr. Marzella	Dr. Marzella requested telecon for 5/25 to discuss AE files and a second is requested to address the IIS protocol
05/23/00	Memo to Dr. Marzella from Dr. Giles	Follow up with Dr. Marzella regarding programming teleconference for safety data
05/25/00	Letter to FDA	Fax to Louis Marzella of FDA containing the adverse event reporting procedures for clinical research and the reporting conventions
05/25/00	Memo to Marzella/Gress (FDA) from Steiner	Provide guidance and written instructions for the use of JMP with the SAS files to run AE tables and data manipulations

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
05/26/00	Letter to FDA	Peg Intron JMP instructions provided to Dr. Marzella of FDA via fax
06/02/00	Letter to FDA	Preclinical monkey study report, SN 99331 (2 books)
06/06/00	Letter to FDA	Desk copy of word 6.0 version c/197-010 narrative
06/07/00	Letter from FDA	Outstanding balance for user fees submitted as new fee schedule put out after submission
06/07/00	BLA Amendment/Letter to FDA	Revised labeling to include "bucket" dosing, complete analysis, Rebetron as 1st line therapy
06/20/00	Memo to Debra Trout (FDA) from Diane Zezza	List of documents to be available for Peg-Intron pre-approval inspection for June 2000
06/23/00	Memo to V. Tyson-Medlock (FDA) from Rachel Steiner	Request for info on pediatric study waiver and CBER system upgrade
07/02/00	BLA Amendment/Letter to FDA	Drug Product attachments: Method of Manufacture; Drug Substance Attachments: Raw materials used in synthesis; Establishments licenses: Water systems, Air Systems
07/18/00	Memo to R. Steiner from Dr. Schwieterman(FDA)	Summary of Peg Monotherapy pediatrics telecon
07/20/00	Letter to FDA	Fax to agency – list of Schering attendees from July 18, 2000 pediatric telecom
07/27/00	Memo to P. Giles from J. Gress (FDA)	Peg Intron Additional data analysis
08/03/00	Memo to Ms. Steiner from Ms. Gress (FDA)	Scheduling of Peg Monotherapy telecon for additional analyses (primarily safety) to be requested
08/04/00	Memo to Steiner from Gress (FDA)	Telecon to discuss additional analyses to be performed for Peg monotherapy submission
08/09/00	Memo to D. Trout/J. Bekisz from D. Zezza	Follow-up on Brinny 483 responses to Peg-IFN pre-approval inspection submitted July 18, 2000
08/09/00	Letter to FDA	Fax to Janet Gress of FDA outlining information to be provided as per the August 4, 2000 teleconference
08/15/00	Letter to FDA	Response to commitments made in 8/9/00 fax – provide additional AE analysis

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
08/16/00	Memo to Rachel Steiner from Dr. Marzella (FDA)	Dr. Marzella needed to know where to find SNA data in the BLA
08/16/00	Memo to R. Steiner from A.Wright (FDA)	Request for C/193-007 protocol
08/17/00	Letter to FDA	Final Analysis requested during 8/9 telecon by Janet Gress; listing of all AES for patient having a serious AE
08/17/00	Memo to V. Tyson-Medlock (FDA) from R. Steiner	PEG action latter date is now 11/13/00 and STN number 103949/0
08/23/00	Memo to R. Steiner from L. Marzella (FDA)	Telecon to discuss c/193-007 labeling change, a plastic anemia safety change, and IIS
08/28/00	Memo to Stuart Feldman from Debbie Trout (FDA)	Peg-Intron PAI – Responses to FDA 483 observations
08/30/00	Letter to FDA	Fax to Anita O'Connor containing preclinical tox. Information for Peg-Intron
08/30/00	Letter to FDA	Fax to Anita O'Connor – pages from investigator's brochure outlining all Peg-Intron Phase I-III studies by indication
08/31/00	Memo to R. Steiner from Vicki Tyson-Medlock (FDA)	Comments on Peg-Intron cartons, Vial and Diluent labeling
08/31/00	Letter to FDA	Fax to Anita O'Connor – pages from PEG BLA, conversion of Peg to Intron doses
08/31/00	Letter to FDA	Fax with comments on diluent, vial, and carton labeling for Peg-Monotherapy application
09/06/00	Memo to Diane Zezza from J. Bekisz (FDA)	Clarification and photographs requested for SDS-Page submission
09/07/00	Letter to FDA	"Bucket" Dosing Proposal Faxed to J.Gress (FDA)
09/07/00	Memo to Rachel Steiner from Glen Jones (FDA)	Summary of FDA telecon on bucket dosing proposal and product reviewer comments (diluent, overfill and labeling issues)
09/08/00	BLA Amendment/Letter to FDA	Bulk sterility test results for Peg Intron and Intron A from 1995-2000. Data requested by Ms. Debbie Trout/office of compliance as a follow-up to our PAI 483 responses

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
09/08/00	Letter to FDA	Fax to FDA with attendee lists for 9/7/00 2 p.m. and 4 p.m. telecons (IIS Peg/Riba study and Peg BLA monotherapy labeling)
09/15/00	Memo to Stuart Feldman from Dr. Joe Bekisz (FDA)	Request for Peg Intron finished product vials, diluent, syringes and needles for demonstration study
09/18/00	Memo to Rachel Steiner from V. Tyson-Medlock (FDA)	Agency follow-up on pediatric proposal for peg monotherapy
09/18/00	Memo to R. Steiner from L. Marzella(FDA)	Agency to follow-up on Peg simplified dosing regression analysis, request for Intron information on hypotension and confirmation
09/18/00	Memo to R.Steiner from P. Haseman(FDA)	Agency requested "protocol deviations" section of study report
09/19/00	Letter from FDA	Request for designation filing has been accepted. Jurisdiction determination for Peg-Intron pen will be issued on November 20, 2000
09/20/00	Letter to FDA	Fax to Dr. Marzella – regarding regression analysis requested by Dr. David Green during 9/7/00 teleconference
09/27/00	Memo to L. Marzella(FDA) from R. Steiner	Confirmation regression analysis fax was received by FDA, request for minor changes to the dosing table (Change weight cuts so there is no overlap in KG. Column, add column to provide actual gram dose to be administered)
09/27/00	Letter to FDA	Peg Intron Logo for preclearance
09/29/00	Letter to FDA	Response to 8/31/00 fax from V. Tyson-Medlock and 9/7/00 telecon issues – revised vial, carton, diluent, labeling, APP manufacturing information regression analysis
10/02/00	Memo to P. Giles from L. Marzella(FDA)	Informal request for additional safety analyses
10/03/00	Memo to Dr. Joe Bekisz (FDA) from Stuart Feldman	Shearwater DMF for SC-PEG
10/04/00	BLA Amendment/Letter to FDA	Biological Variance notification (BVN) and investigation report on bulk sterility failure for Intron MD Pen Batch 0-IOL-123 –Follow up request to September 8, 2000 Amendment

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
10/04/00	Letter to FDA	Letter to Mr. James Hamilton (Import/Export International Team) requesting his assistance in obtaining drug listing for Peg-Intron prior to BLA approval (early importation required for product launch).
10/04/00	Memo to Dr. Joe Bekisz (FDA) from Stuart Feldman	Shearwater DMF for SC-PEG
10/04/00	Memo to Debbie Trout (FDA) from Stuart Feldman	Peg-Intron PAI – Status of September 8 response.
10/04/00	Letter to FDA	Additional 4 copies of Schering's 9/29/00 submission provided (response to 8/31/00 fax and 9/7/00 telecom issues).
10/04/00	Memot o Ms. Debbie Trout (FDA) from Stuart Feldman	BVN and investigation report on bulk sterility failure for pen batch 0-I0l-123. This was follow-up request from FDA to our September 8, 2000 submission (bulk sterility test results). Note: The 9/8 submission was a follow-up to our PAI responses submitted on 7/18/00.
10/06/00	Memo to Dr. Joe Bekisz (FDA) from Stuart Feldman	Shearwater DMF for SC-PEG
10/06/00	Letter to FDA	Fax to Janet Gress of FDA discussing incidence/duration/severity of serious adverse events occurring in responders vs. relapsers vs. nonresponders.
10/09/00	Letter to FDA	Submission of revised AE listings per Dr. Marcella's request.
10/10/00	Memo to Jim Hamilton (FDA) from Stuart Feldman	Importation of Peg-Intron
10/11/00	Memo to Joe Bekisz (FDA) from Stuart Feldman	List of Schering attendees for October 1, 2000 telecom Re: SC-PEG
10/12/00	Memo to Dr. Stuart Feldman from Dr. Bekisz/Dr. Dye/Dr. Nyugen (FDA)	Peg Intron PAI – 483 responses

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
10/12/00	Memo to Dr. Stuart Feldman from Dr. Bekisz/Dr. Dye/Dr. Nyugen (FDA	Drug substance reprocessing and drug product overfill.
10/12/00	Memo to Dye, Beaucage, Nagle, Voloch (FDA) from Feldman	SC-PEG starting material
10/18/00	Letter to FDA	Fax to Dr. Marcella of FDA containing revised AE table and request PEG and Intron clin. Pharm values (T ½ and CL/F).
10/19/00	Letter to FDA	Hard copy of information provided to Dr. Marcella via fax 10/18/00.
10/19/00	Memo to R. Steiner from V. Tyson-Medlock (FDA)	Comments on the vial labels, diluent label and carton labels for Peg Intron.
10/20/00	Letter to FDA	Fax follow-up for preclearance of Peg-Intron logo sent September 27, 2000.
10/23/00	Memo to Dr. Joe Bekisz (FDA) from Dr. Stuart Feldman	Faxed copy of amendment submitted on 10/23/00.
10/23/00	BLA Amendment	CMC information requested by CBER: 1) status report on Shearwater DMF deficiency letter and 2) revised overfill justification report.
10/24/00	Letter from FDA	Proposed PEG-Intron logo – add "Powder" before "For Injection".
10/27/00	Letter to FDA	Pediatric infor request for monotherapy waiver.
10/30/00	Memo to W. Schwieterman (FDA) from P. Giles	Dr. Schwieterman agreed to follow-up on the status of the Peg-Monotherapy labeling comments and reiterated that for the Peg-Riba meeting, 24 week follow-up data must be in the briefing book.
10/31/00	Memo to L. Marzella (FDA) from Giles/Steiner	Preliminary comments on Peg Monotherapy product info sheet.
11/01/00	Letter to FDA	Fax to V. Tyson of FDA containing copies of PEG Intron carton sample for comment.
11/03/00	Letter to FDA	Pak 3 Intron a solution box sent as desk copy to V. Tyson-Medlock of FDA.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
11/03/00	Memo to Rachael Steiner from Vicky Tyson-Medlock (FDA)	Comments on the diluent label and carton labels for PEG-Intron.
11/06/00	Letter to FDA	Alcohol swab and syringe information.
11/06/00	Memo to W. Schwieterman (FDA) from P. Giles	Discussion of issues for approval of PEG monotherapy application: Patient med guide, last minute changes outside the div. and SAE/class labeling for interferons.
11/07/00	Letter to FDA	Revised diluent, vial and carton labels incorporating all FDA comments and syringe/alcohol swab information.
11/13/00	Memo to B. Schwieterman (FDA) from P. Giles	FDA intends to send complete review letter for Peg Monotherapy BLA, labeling and Phase IV commitments being worked out.
11/13/00	Letter from FDA	Complete review letter for Peg Monotherapy received.
11/13/00	Letter from FDA	FDA's version of the Peg-Intron monotherapy label.
11/16/00	Memo to Dr. Schwieterman (FDA) from P. Giles	Schedule telecon to clarify Phase IV commitment requests.
11/17/00	Letter to FDA	Intent to file amendment in response to Nov. 13, 2000 approvable letter.
11/17/00	Memo to Vicky Tyson-Medlock (FDA) from Rachael Steiner	Storage conditions.
11/20/00	Letter from FDA	Approval for request for designation for PEG-Intron pen was accepted by FDA. CBER will be the agency division with primary review responsibility for the en. (2000.19)
11/22/00	Memo to Schwieterman, Greene, Marzella from Giles	Discussion on Phase IV commitments.
11/28/00	Memo to Dr. Schwieterman (FDA) from P. Giles	Labeling.
11/28/00	Letter to FDA	Response to complete review/approvable letter dated November 13, 2000.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
12/01/00	Memo to Rachael Steiner from V. Tyson-Medlock (FDA)	Telecon set up for Peg Intron monotherapy labeling.
12/05/00	Memo to Rachael Steiner from V. Tyson-Medlock (FDA)	Telecon on Labeling.
12/05/00	Letter to FDA	Final draft carton and vial labeling for Peg Intron.
12/06/00	Memo to Penny Giles from Vicky Tyson-Medlock (FDA)	Peg Intron labeling.
12/07/00	Memo to Rachael Steiner from Louis Marzella (FDA)	Response to complete response letter.
12/11/00	Letter from FDA	Complete response letter received and acceptable. FDA to respond by Jan. 29, 2001 (BL 103949/0).
12/12/00	Memo to Dr. Schwieterman (FDA) from Penny Giles	Labeling and pre-BLA meeting PEG and Riba.
12/13/00	Letter from FDA	Peg Intron FDA proposed labeling.
12/20/00	Memo supplement to L. Marzella (FDA) from P. Giles	Schedule telecon to discuss African American Study.
12/21/00	Memo supplement to R. Steiner from V. Tyson-Medlock (FDA)	If med guide is required, additional changes will be needed to the carton text.
12/21/00	Memo to Steiner/Giles/ Lamendola	Phase IV commitments.
12/22/00	Letter to FDA	Schering's study plans to evaluate non-caucasians-study will use Peg Triba (103949).
01/05/01	Memo to Penny Giles from Bill Schwieterman (FDA)	Peg mono review status and P/R electronic files for adverse events.
01/08/01	Letter from FDA	Fax from FDA to revise storage statement to include excursions.
01/10/01	Letter from FDA	Peg-Intron PI and med guide sent via e-mail from CBER to Schering.
01/11/01	Letter to FDA	Fax to FDA of cover letter from 12/20/00 labeling submission.
01/11/01	Letter from FDA	January 10, 2001 versions of package insert and medication guide proposed by FDA.

DOCUMENT DATE	DOCUMENT TYPE	SUBJECT
01/12/01	Letter to FDA	Final draft labeling submitted fro Peg-Intron monotherapy (PI, Med Guide, Product Vial Labels, Cartons).
01/12/01	Letter to FDA	Timing for the Phase IV Peg-Intron mandatory Phase 4 commitment studies to be performed submitted.
01/17/01	Memo to J. Lamendola from Glen Jones (FDA)	Discussion: status of Peg and Intron BLA approval.
01/17/01	Letter from FDA	Fax from FDA noting requested changes on the cartons.
01/18/01	Letter to FDA	Draft press release and CMC supplement for water/diluent to be submitted 12/01.
01/18/01	Letter from FDA	Fax from FDA with Peg Intron package insert.
01/19/01	Letter to FDA	Fax of mocked up carton containing changes and lightened for legibility.
01/19/01	Letter from FDA	E-mail from FDA providing their proposed medication guide electronically.
01/19/01	Letter to FDA	Two faxes to FDA containing the carton and labeling for Peg-Intron.
01/19/01	Letter to FDA supplement	Two faxes containing mocked up labeling for Peg Intron – vials, cartons, PI and medication guide.
01/19/01	Approval letter from FDA	Approval letter for Peg Monotherapy, 5 Phase IV commitments listed and press release with FDA's comments provided.
01/22/01	Letter to FDA	Fax to FDA with mocked up cartons for the 100, 160, 300 ug/ml. Strengths for Peg Intron.
01/22/01	Letter to FDA	Faxes and final draft labeling for the BLA submitted (103949)
01/22/01	Letter from FDA	Fax from FDA – Form 2253 for the Peg Intron Monotherapy press release.

PEG-Intron™
(Peginterferon alfa-2b)
Powder for Injection

EXHIBIT XII

WARNING
Alpha interferons, including PEG-Intron, cause or aggravate fatal or life-threatening neuropsychiatric, autoimmune, ischemic, and infectious disorders. Patients should be monitored closely with periodic clinical and laboratory evaluations. Patients with persistently severe or worsening signs or symptoms of these conditions should be withdrawn from therapy. In many but not all cases these disorders resolve after stopping PEG-Intron therapy. See **WARNINGS, ADVERSE REACTIONS**.

DESCRIPTION
PEG-Intron™, peginterferon alfa-2b Powder for Injection, is a covalent conjugate of recombinant alfa interferon with monomethoxy polyethylene glycol (PEG). The molecular weight of the PEG portion of the molecule is 12,000 daltons. The average molecular weight of the PEG-Intron molecule is approximately 31,000 daltons. The specific activity of pegylated interferon alfa-2b is approximately 0.7 x 10^8 IU/mg protein.

Interferon alfa-2b, the starting material used to manufacture PEG-Intron, is a water-soluble protein with a molecular weight of 19,271 daltons produced by recombinant DNA techniques. It is obtained from the bacterial fermentation of a strain of *Escherichia coli* bearing a genetically engineered plasmid containing an interferon gene from human leukocytes.

PEG-Intron is a white to off-white lyophilized powder supplied in 2-mL vials for subcutaneous use. Each vial contains either 74 µg, 118.4 µg, 177.6 µg, or 222 µg of PEG-Intron, and 1.11 mg dibasic sodium phosphate anhydrous, 1.11 mg monobasic sodium phosphate dihydrate, 59.2 mg sucrose and 0.074 mg polysorbate 80. Following reconstitution with 0.7 mL of the supplied diluent (Sterile Water for Injection, USP), each vial contains PEG-Intron at strengths of either 100 µg/mL, 160 µg/mL, 240 µg/mL, or 300 µg/mL.

CLINICAL PHARMACOLOGY
General: The biological activity of PEG-Intron is derived from its interferon alfa-2b moiety. Interferons exert their cellular activities by binding to specific membrane receptors on the cell surface and initiate a complex sequence of intracellular events. These include the induction of certain enzymes, suppression of cell proliferation, immunomodulating activities such as enhancement of the phagocytic activity of macrophages and augmentation of the specific cytotoxicity of lymphocytes for target cells, and inhibition of virus replication in virus-infected cells. Interferon alfa upregulates the Th1 T-helper cell subset in *in vitro* studies. The clinical relevance of these findings is not known.

Pharmacodynamics: PEG-Intron raises concentrations of effector proteins such as serum neopterin and 2′5′ oligoadenylate synthetase, raises body temperature, and causes reversible decreases in leukocyte and platelet counts. The correlation between the *in vitro* and *in vivo* pharmacologic and pharmacodynamic and clinical effects is unknown.

Pharmacokinetics: Following a single subcutaneous dose of PEG-Intron, the mean absorption half-life (t $1/2$ k_a) was 4.6 hours. Maximal serum concentrations (C_{max}) occur between 15-44 hours post-dose, and are sustained for up to 48-72 hours. The C_{max} and AUC measurements of PEG-Intron increase in a dose-related manner. After multiple dosing, there is an increase in bioavailability of PEG-Intron. Week 48 mean trough concentrations (320 pg/mL; range 0, 2960) are approximately 3-fold higher than Week 4 mean trough concentrations (94 pg/mL; range 0, 416). The mean PEG-Intron elimination half-life is approximately 40 hours (range 22 to 60 hours) in patients with HCV infection. The apparent clearance of PEG-Intron is estimated to be approximately 22.0 mL/hr•kg. Renal elimination accounts for 30% of the clearance. Single dose peginterferon alfa-2b pharmacokinetics following a subcutaneous 1.0 µg/kg dose suggest the clearance of peginterferon alfa-2b is reduced by approximately half in patients with impaired renal function (creatinine clearance <50 mL/minute).

Pegylation of interferon alfa-2b produces a product (PEG-Intron) whose clearance is lower than that of non-pegylated interferon alfa-2b. When compared to INTRON A, PEG-Intron (1.0 µg/kg) has approximately a seven-fold lower mean apparent clearance and a five-fold greater mean half-life permitting a reduced dosing frequency. At effective therapeutic doses, PEG-Intron has approximately ten-fold greater C_{max} and 50-fold greater AUC than interferon alfa-2b.

Pharmacokinetic data from geriatric patients (> 65 years of age) treated with a single subcutaneous dose of 1.0 µg/kg of PEG-Intron showed no remarkable differences in C_{max}, AUC, clearance, or elimination half-life from those obtained in younger patients.

During the 48 week treatment period with PEG-Intron no differences in the pharmacokinetic profiles were observed between male and female patients with chronic hepatitis C infection.

Drug Interactions: It is not known if PEG-Intron therapy causes clinically significant drug-drug interactions with drugs metabolized by the liver in patients with hepatitis C. In 12 healthy subjects known to be CYP2D6 extensive metabolizers, a single subcutaneous dose of 1 µg/kg PEG-Intron did not inhibit CYP1A2, 2C8/9, 2D6, hepatic 3A4 or N-acetyltransferase; the effects of PEG-Intron on CYP2C19 were not assessed.

CLINICAL STUDIES
A randomized study compared treatment with PEG-Intron (0.5, 1.0, or 1.5 µg/kg once weekly SC) to treatment with INTRON A, (3 million units three times weekly SC) in 1219 adults with chronic hepatitis from HCV infection. The patients were not previously treated with interferon alfa, had compensated liver disease, detectable HCV RNA, elevated ALT, and liver histopathology consistent with chronic hepatitis. Patients were treated for 48 weeks and were followed for 24 weeks post-treatment. Seventy percent of all patients were infected with HCV genotype 1, and 74% of all patients had high baseline levels of HCV RNA (more than 2 million copies per mL of serum), two factors known to predict poor response to treatment.

Response to treatment was defined as undetectable HCV RNA and normalization of ALT at 24 weeks post-treatment. The response rates to the 1.0 and 1.5 µg/kg PEG-Intron doses were similar to each other and were both higher than response rates to INTRON A. (See **Table 1**)

TABLE 1. Rates of Response to Treatment

	A PEG-Intron 0.5 µg/kg (N=315)	B PEG-Intron 1.0 µg/kg (N=298)	C INTRON A 3 MIU TIW (N=307)	B-C (95% CI) Difference between PEG-Intron 1.0 ug/kg and INTRON A
Treatment Response (Combined Virologic Response and ALT Normalization)	17%	24%	12%	11 (5, 18)
Virologic Response*	18%	25%	12%	12 (6, 9)
ALT Normalization	24%	29%	18%	11 (5, 18)

*Serum HCV RNA is measured by a research-based quantitive polymerase chain reaction with a lower limit of detection of 100 copies/mL at the National Genetics Institute, Culver City, CA.

Patients with both viral genotype 1 and high serum levels of HCV RNA at baseline were less likely to respond to treatment with PEG-Intron. Among patients with the two unfavorable prognostic variables, 8% (12/157) responded to PEG-Intron treatment and 2% (4/169) responded to INTRON A. Doses of PEG-Intron higher than the recommended dose did not result in higher response rates in these patients.

Patients receiving PEG-Intron with viral genotype 1 had a response rate of 14% (28/199) while patients with other viral genotypes had a 45% (43/96) response rate.

Ninety-six percent of the responders in the PEG-Intron groups and 100% of responders in the INTRON A group first cleared their viral RNA by week 24 of treatment. See **DOSAGE AND ADMINISTRATION**.

The treatment response rates were similar in men and women. Response rates were lower in African American and Hispanic patients and higher in Asians compared to Caucasians. Although African Americans had a higher proportion of poor prognostic factors compared to Caucasians the number of non-Caucasians studied (9% of the total) was insufficient to allow meaningful conclusions about differences in response rates after adjusting for prognostic factors.

Liver biopsies were obtained before and after treatment in 60% of patients. A modest reduction in inflammation compared to baseline that was similar in all four treatment groups was observed.

INDICATIONS AND USAGE

PEG-Intron, peginterferon alfa-2b, monotherapy is indicated for the treatment of chronic hepatitis C in patients not previously treated with interferon alpha who have compensated liver disease and are at least 18 years of age. The safety and efficacy of peginterferon alfa-2b (PEG-Intron) in combination with ribavirin (REBETOL) for the treatment of chronic hepatitis C have not been established.

CONTRAINDICATIONS

PEG-Intron is contraindicated in patients with:

- hypersensitivity to PEG-Intron or any component of the product
- autoimmune hepatitis
- decompensated liver disease

WARNINGS

Patients should be monitored for the following serious conditions, some of which may become life threatening. Patients with persistently severe or worsening signs or symptoms should be withdrawn from therapy.

Neuropsychiatric events

Life-threatening or fatal neuropsychiatric events, including suicide, suicidal and homicidal ideation, depression, relapse of drug addiction/overdose, and aggressive behavior have occurred in patients with and without a previous psychiatric disorder during PEG-Intron treatment and follow-up. Psychoses and hallucinations have been observed in patients treated with alpha interferons. PEG-Intron should be used with extreme caution in patients with a history of psychiatric disorders. Patients should be advised to report immediately any symptoms of depression and/or suicidal ideation to their prescribing physicians. Physicians should monitor all patients for evidence of depression and other psychiatric symptoms. In severe cases, PEG-Intron should be stopped immediately and psychiatric intervention instituted.

Bone marrow toxicity

PEG-Intron suppresses bone marrow function, sometimes resulting in severe cytopenias. PEG-Intron should be discontinued in patients who develop severe decreases in neutrophil or platelet counts. Very rarely alpha interferons may be associated with aplastic anemia. (See **DOSAGE AND ADMINISTRATION**)

Endocrine disorders

PEG-Intron causes or aggravates hypothyroidism and hyperthyroidism. Hyperglycemia has been observed in patients treated with PEG-Intron. Diabetes mellitus has been observed in patients treated with alpha interferons. Patients with these conditions who cannot be effectively treated by medication should not begin PEG-Intron therapy. Patients who develop these conditions during treatment and cannot be controlled with medication should not continue PEG-Intron therapy.

Cardiovascular events

Cardiovascular events, which include hypotension, arrhythmia, tachycardia, cardiomyopathy, and myocardial infarction have been observed in patients treated with PEG-Intron. PEG-Intron should be used cautiously in patients with cardiovascular disease. Patients with a history of myocardial infarction and arrhythmic disorder who require PEG-Intron therapy should be closely monitored (see **Laboratory tests**).

Colitis

Fatal and nonfatal ulcerative and hemorrhagic colitis has been observed within 12 weeks of the start of alpha interferon treatment. Abdominal pain, bloody diarrhea, and fever are the typical manifestations. PEG-Intron treatment should be discontinued immediately in patients who develop these symptoms and signs. The colitis usually resolves within 1-3 weeks of discontinuation of alpha interferons.

Pancreatitis

Fatal and nonfatal pancreatitis has been observed in patients treated with alpha interferon. PEG-Intron therapy should be suspended in patients with signs and symptoms suggestive of pancreatitis and discontinued in patients diagnosed with pancreatitis.

Autoimmune disorders

Development or exacerbation of autoimmune disorders (e.g., thyroiditis, thrombocytopenia, rheumatoid arthritis, interstitial nephritis, systemic lupus erythematosus, psoriasis) have been observed in patients receiving PEG-Intron. PEG-Intron should be used with caution in patients with autoimmune disorders.

Pulmonary disorders

Dyspnea, pulmonary infiltrates, pneumonitis and pneumonia, some resulting in patient deaths, have been associated with PEG-Intron or alpha interferon therapy. Patients with pulmonary infiltrates or pulmonary function impairment should be closely monitored.

Hypersensitivity

Serious, acute hypersensitivity reactions (e.g., urticaria, angioedema, bronchoconstriction, anaphylaxis) have been rarely observed during alpha interferon therapy. If such a reaction develops during treatment with PEG-Intron, discontinue treatment and institute appropriate medical therapy immediately. Transient rashes do not necessitate interruption of treatment.

PRECAUTIONS

- PEG-Intron has not been studied in patients who have failed other alpha interferon treatments.
- The safety and efficacy of PEG-Intron for the treatment of hepatitis C in patients who have received liver or other organ transplant recipients have not been studied.
- The safety and efficacy of PEG-Intron for the treatment of patients with HCV coinfected with HIV or HBV have not been established.

Ophthalmologic disorders: Retinal hemorrhages, cotton wool spots, and retinal artery or vein obstruction have been observed after treatment with PEG-Intron or alpha interferons. Patients who have diabetes mellitus or hypertension should have eye examinations before the start of PEG-Intron treatment.

Patients with renal failure: Patients with impairment of renal function should be closely monitored for signs and symptoms of interferon toxicity and doses of PEG-Intron should be adjusted accordingly. PEG-Intron should be used with caution in patients with creatinine clearance <50 mL/min. See **DOSAGE AND ADMINISTRATION.**

Immunogenicity: One percent of patients (7/734) receiving PEG-Intron developed low-titer (≤64) neutralizing antibodies to INTRON A. The clinical and pathological significance of the appearance of serum neutralizing antibodies is unknown. No apparent correlation of antibody development to clinical response or adverse events was observed. The incidence of post-treatment binding antibody was approximately 10% for patients receiving PEG-Intron and approximately 15% for patients receiving INTRON A. The data reflect the percentage of patients whose test results were considered positive for antibodies to PEG-Intron in a Biacore assay that is used to measure binding antibodies, and in an antiviral neutralization assay which measures serum neutralizing antibodies. The percentage of patients whose test results were considered positive for antibodies is highly dependent on the sensitivity and specificity of the assays. Additionally the observed incidence of antibody positivity in these assays may be influenced by several factors including sample timing and handling, concomitant medications, and underlying disease. For these reasons, comparison of the incidence of antibodies to PEG-Intron with the incidence of antibodies to other products may be misleading.

- **Laboratory Tests:** PEG-Intron may cause severe decreases in neutrophil and platelet counts, and abnormality of TSH. In 10% of patients treated with PEG-Intron ALT levels rose 2 to 5-fold above baseline. The elevations were transient and were not associated with deterioration of other liver functions.

Patients on PEG-Intron therapy should have hematology and blood chemistry testing before the start of treatment and then periodically thereafter. In the clinical trial CBC (including neutrophil and platelet counts) and chemistries (including AST, ALT, and bilirubin) were measured during the treatment period at weeks 2, 4, 8, 12, and then at 6-week intervals or more frequently if abnormalities developed. TSH levels were measured every 12 weeks during the treatment period.

Patients who have pre-existing cardiac abnormalities should have electrocardiograms administered before treatment with PEG-Intron.

Information for Patients: Patients receiving PEG-Intron should be directed in its appropriate use, informed of the benefits and risks associated with treatment, and referred to the **MEDICATION GUIDE.**

A puncture-resistant container for the disposal of used syringes and needles should be supplied to the patient for at home use. Patients should be thoroughly instructed in the importance of proper disposal and cautioned against any reuse of needles and syringes. The full container should be disposed of according to the directions provided by the physician (see **MEDICATION GUIDE**).

Patients should be informed that there are no data evaluating whether PEG-Intron therapy will prevent transmission of HCV infection to others. Also, it is not known if treatment with PEG-Intron will cure hepatitis C or prevent cirrhosis, liver failure, or liver cancer that may be the result of infection with the hepatitis C virus.

Patients should be advised that laboratory evaluations are required before starting therapy and periodically thereafter (see **Laboratory Tests**). It is advised that patients be well-hydrated, especially during the initial stages of treatment. "Flu-like" symptoms associated with administration of PEG-Intron may be minimized by bedtime administration of PEG-Intron or by use of antipyretics.

Carcinogenesis, Mutagenesis, Impairment of Fertility

Carcinogenesis: PEG-Intron has not been tested for its carcinogenic potential.

Mutagenesis: Neither PEG-Intron, nor its components interferon or methoxypolyethylene glycol caused damage to DNA when tested in the standard battery of mutagenesis assays, in the presence and absence of metabolic activation.

Impairment of Fertility: Irregular menstrual cycles were observed in female cynomolgus monkeys given subcutaneous injections of 4239 μg/m² PEG-Intron every other day for one month, at approximately 345 times the recommended weekly human dose (based upon body surface area). These effects included transiently decreased serum levels of estradiol and progesterone, suggestive of anovulation. Normal menstrual cycles and serum hormone levels resumed in these animals 2 to 3 months following cessation of PEG-Intron treatment. Every other day

dosing with 262 µg/m² (approximately 21 times the weekly human dose) had no effects on cycle duration or reproductive hormone status. The effects of PEG-Intron on male fertility have not been studied.

Pregnancy Category C: Non-pegylated Interferon alfa-2b, has been shown to have abortifacient effects in *Macaca mulatta* (rhesus monkeys) at 15 and 30 million IU/kg (estimated human equivalent of 5 and 10 million IU/kg, based on body surface area adjustment for a 60 kg adult). PEG-Intron should be assumed to also have abortifacient potential. There are no adequate and well-controlled studies in pregnant women. PEG-Intron therapy is to be used during pregnancy only if the potential benefit justifies the potential risk to the fetus. Therefore, PEG-Intron is recommended for use in fertile women only when they are using effective contraception during the treatment period.

Nursing Mothers: It is not known whether the components of PEG-Intron are excreted in human milk. Because of the potential for adverse reactions from the drug in nursing infants, a decision must be made whether to discontinue nursing or discontinue the treatment, taking into account the importance of the product to the mother.

Pediatric Use Safety and effectiveness in pediatric patients below the age of 18 years have not been established.

Geriatric Patients Clinical studies of PEG-Intron did not include sufficient numbers of subjects aged 65 and over to determine whether they respond differently than younger subjects. Other reported clinical experience has not identified differences in responses between the elderly and younger patients. However, treatment with alpha interferons, including PEG-Intron, is associated with CNS, cardiac, and systemic (flu-like) adverse effects. Because these adverse reactions may be more severe in the elderly, caution should be exercised in use of PEG-Intron in this population. This drug is known to be substantially excreted by the kidney. Because elderly patients are more likely to have decreased renal function, the risk of toxic reactions to this drug may be greater in patients with impaired renal function.

ADVERSE REACTIONS

Nearly all study patients experienced one or more adverse events. The incidence of serious adverse events was similar (about 12%) in all treatment groups. In many but not all cases, events resolve after stopping PEG-Intron therapy. Some patients continued to experience adverse events for several months after discontinuation of therapy. There was one patient death, a suicide, among patients receiving PEG-Intron and two patient deaths in the INTRON A group (1 murder/suicide and 1 sudden death). Overall, 10% of patients in the PEG-Intron groups discontinued therapy due to adverse events compared to 6% in the INTRON A group. Fourteen percent of patients in the PEG-Intron groups required dose reduction compared to 6% in the INTRON A group.

The most common adverse events associated with PEG-Intron were "flu-like" symptoms which occurred in approximately 50% of patients, and may decrease in severity as treatment continues. Application site disorders occurred frequently (47%) and included injection site inflammation, and reaction (i.e. bruise, itchiness, irritation). Injection site pain was reported in 2% of patients receiving PEG-Intron. Alopecia (thinning of the hair) is also often associated with PEG-Intron.

Fifty-seven percent of patients treated with PEG-Intron experienced psychiatric adverse events, most commonly depression (29%). Suicidal behavior (ideation, attempts, and suicides) occurred in 1% of all patients during or shortly after treatment with PEG-Intron. (See **WARNINGS**).

Patients receiving PEG-Intron appeared to experience a greater number of adverse events (e.g., injection site reaction, fever, rigors, nausea) compared to patients receiving INTRON A. The number of adverse events in all body systems in general was higher in patients receiving the higher PEG-Intron dosages.

Adverse events that occurred in the Phase 3 clinical trial at ≥5% incidence are provided in **Table 2** by treatment group.

TABLE 2. Adverse Events Occurring in ≥5% of Patients

Adverse Events	PEG-Intron 1.0 µg/kg (N=297)	INTRON A 3 MIU (N=303)
	Percentage of Patients Reporting Adverse Events*	
Application Site Disorders		
Injection Site Inflammation/Reaction	47	20
Autonomic Nervous System Disorders		
Flushing	6	3
Sweating Increased	6	7
Body as a Whole –General Disorders		
Headache	56	52
Fatigue	52	54
Influenza-Like Symptoms	46	38
Rigors	23	19
Fever	22	12
Weight Decrease	11	13
Body as a Whole –General Disorders *cont'd.*		
RUQ pain	8	8
Malaise	7	6
Central and Peripheral Nervous System Disorders		
Dizziness	12	10
Hypertonia	5	3
Endocrine Disorders		
Hypothyrodism	5	3
Gastro-Intestinal System Disorders		
Nausea	26	20
Anorexia	20	17
Diarrhea	18	16
Abdominal Pain	15	11
Vomiting	7	6
Dyspepsia	6	7
Hemotologic Disorders		
Neutropenia	6	2
Thrombocytopenia	7	<1
Infectious Disorders		
Infection Viral	11	10
Liver and Biliary System Disorders		
Hepatomegaly	6	5
Musculoskeletal System Disorders		
Musculoskeletal Pain	56	58
Psychiatric Disorders		
Depression	29	25
Insomnia	23	23
Anxiety/Emotional Lability/Irritability	28	34
Respiratory System Disorders		
Pharyngitis	10	7
Sinusitis	7	7
Coughing	6	5
Skin and Appendages Disorders		
Alopecia	22	22
Pruritus	12	8
Dry skin	11	9
Rash	6	7

* Patients reporting one or more adverse events. A patient may have reported more than one adverse event within a body system/organ class category.

Numerous adverse events were observed at a frequency <5%. In the absence of a non-treatment control group the relationship to study drug could not be determined.

Individual serious adverse events occurred at a frequency ≤1% and included suicide attempt, suicidal ideation, severe depression; relapse of drug addiction/overdose; nerve palsy (facial, oculomotor); cardiomyopathy, myocardial infarction, retinal ischemia, retinal vein thrombosis, transient ischemic attack, supraventricular arrhythmias, loss of consciousness; neutropenia, infection (pneumonia, abscess); autoimmune thrombocytopenia, hyperthyroidism, rheumatoid arthritis, interstitial nephritis, lupus-like syndrome, aggravated psoriasis; urticaria.

Laboratory Values

Neutrophils Neutrophil counts decreased in 70% of patients. Severe potentially life-threatening neutropenia (<0.5 x 10^9/L) occurred in 1% of patients.

Platelets Platelet counts decreased in 20% of patients. Treatment with PEG-Intron resulted in severe decreases in platelet counts (<50,000/mm³) in 1% of patients.

The incidence and severity of thrombocytopenia and neutropenia were greater in the PEG-Intron groups compared to the interferon alfa group. Platelet and neutrophil counts generally returned to pretreatment levels within 4 weeks of the cessation of therapy.

Thyroid Function TSH abnormalities developed in 16% of patients and were associated with clinically apparent hypothyroidism (5%) or hyperthyroidism (1%). Subjects developed new onset TSH abnormalities while on treatment and during the follow-up period. At the end of the follow-up period 7% of subjects still had abnormal TSH values.

OVERDOSAGE

There is limited experience with overdosage. In the clinical study, 13 patients acci-

dentally received a dose greater than that prescribed. There were no instances in which a patient received more than 2.5 times the intended dose. The maximum dose received by any patient was 3.45 µg/kg weekly over a period of approximately 12 weeks. There were no serious reactions attributed to these overdosages.

DOSAGE AND ADMINISTRATION

A patient should self-inject only if the physician determines that it is appropriate and the patient agrees to medical follow-up as necessary and training in proper injection technique has been given to him/her. (See illustrated **MEDICATION GUIDE** for instructions.)

PEG-Intron is administered subcutaneously once weekly for one year. The dose should be administered on the same day of each week. Initial dosing should be based on weight as described in **Table 3**.

TABLE 3. Recommended Dosing

Vial Strength* to Use (µg/mL)	Weight (kg)	Amount of PEG-Intron to Administer (µg)	Volume of PEG-Intron* to Administer (mL)
100	37-45	40	0.4
	46-56	50	0.5
160	57-72	64	0.4
	73-88	80	0.5
240	89-106	96	0.4
	107-136	120	0.5
300	137-160	150	0.5

* When reconsituted as directed

Serum HCV RNA levels should be assessed after 24 weeks of treatment. Discontinuation of treatment should be considered in any patient who has not achieved an HCV RNA below the limit of detection of the assay after 24 weeks of therapy with PEG-Intron. (See **CLINICAL STUDIES.**)

There are no safety and efficacy data for treatment longer than 48 weeks or for re-treatment of patients who relapse following PEG-Intron therapy.

Dose Reduction

If a serious adverse reaction develops during the course of treatment (see **WARNINGS**) discontinue or modify the dosage of PEG-Intron to one-half the starting dosage until the adverse event abates or decreases in severity. If persistent or recurrent intolerance develops despite adequate dosage adjustment, discontinue treatment with PEG-Intron. For dose modification in the event of neutropenia and thrombocytopenia see **Table 4**.

TABLE 4. Guidelines for Dose Modifications for Neutropenia and Thrombocytopenia

	Dose Reduction	Permanent Discontinuation
Neutrophil Count	$<0.75 \times 10^9$/L	$<0.50 \times 10^9$/L
Platelet Count	$<80 \times 10^9$/L	$<50 \times 10^9$/L

Preparation and Administration

Two B-D Safety Lok™ syringes are provided in the package; one syringe is for the reconstitution steps and one for the patient injection. There is a plastic safety sleeve to be pulled over the needle after use. The syringe locks with an audible click when the green stripe on the safety sleeve covers the red stripe on the needle. Brief instructions for the preparation and administration of PEG-Intron Powder for Injection are provided below. Please refer to the **MEDICATION GUIDE** for detailed, step by step instuctions.

Reconstitute the PEG-Intron lyophilized product with only 0.7 mL of supplied diluent (Sterile Water for Injection, USP). **The diluent vial is for single use only. The remaining diluent should be discarded.** No other medications should be added to solutions containing PEG-Intron, and PEG-Intron should not be reconstituted with other diluents. Swirl gently to hasten complete dissolution of the powder. The reconstituted solution should be clear and colorless. Visually inspect the solution for particulate matter and discoloration prior to administration. The solution should not be used if discolored or cloudy, or if particulates are present. (See **MEDICATION GUIDE** for detailed instructions).

The reconstituted solution should be used immediately and cannot be stored for more than 24 hours at 2°-8°C (see **Storage**). The appropriate PEG-Intron dose should be withdrawn and injected subcutaneously. (See **MEDICATION GUIDE** for detailed instructions.) The PEG-Intron vial is a single use vial and does not contain a preservative. **DO NOT REENTER VIAL. DISCARD UNUSED PORTION.** Once the dose from a single dose vial has been withdrawn, the sterility of any remaining product can no longer be guaranteed. Pooling of unused portions of some medications has been linked to bacterial contamination and morbidity.

After preparation and administration of the PEG-Intron injection, it is essential to follow the procedure for proper disposal of syringes and needles. A puncture-resistant container should be used for disposal of syringes. Patients should be instructed in the technique and importance of proper syringe disposal and be cautioned against reuse of these items (see **MEDICATION GUIDE** for detailed instructions.)

Storage

PEG-Intron, should be stored at 25°C (77°F): excursions permitted to 15-30°C (59-86°F)[see USP Controlled Room Temperature]. After reconstitution with supplied Diluent the solution should be used immediately, but may be stored up to 24 hours at 2° to 8°C (36° to 46°F). The reconstituted solution contains no preservative, is clear and colorless. **Do not freeze.**

HOW SUPPLIED

PEG-Intron is a white to off-white lyophilized powder supplied in 2-mL vials. The PEG-Intron Powder for Injection should be reconstituted with 0.7 mL of the supplied Diluent (Sterile Water for Injection, USP) prior to use.

	Each PEG-Intron Package Contains:	
For Patients 37-56 kg	A box containing one 100 µg/mL vial of PEG-Intron Powder for Injection and one 5 mL vial of Diluent (Sterile Water for Injection, USP), 2 B-D Safety Lok™ syringes with a safety sleeve and 2 alcohol swabs.	(NDC 0085-1368-01)
For Patients 57-88 kg	A box containing one 160 µg/mL vial of PEG-Intron Powder for Injection and one 5 mL vial of Diluent (Sterile Water for Injection, USP), 2 B-D Safety Lok™ syringes with a safety sleeve and 2 alcohol swabs.	(NDC 0085-1291-01)
For Patients 89-136 kg	A box containing one 240 µg/mL vial of PEG-Intron Powder for Injection and one 5 mL vial of Diluent (Sterile Water for Injection, USP), 2 B-D Safety Lok™ syringes with a safety sleeve and 2 alcohol swabs.	(NDC 0085-1304-01)
For Patients 137-160 kg	A box containing one 300 µg/mL vial of PEG-Intron Powder for Injection and one 5 mL vial of Diluent (Sterile Water for Injection, USP), 2 B-D Safety Lok™ syringes with a safety sleeve and 2 alcohol swabs.	(NDC 0085-1279-01)

Schering®

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EXHIBIT XIII

UNITED STATES PATENT AND TRADEMARK OFFICE

CERTIFICATE OF CORRECTION

PATENT NO. : 5,951,974

DATED : September 14, 1999

INVENTOR(S) : GILBERT, et al.

It is certified that error appears in the above-identified patent and that said Letters Patent are hereby corrected as shown below:

Claim 19, line 1: "19" should read –18–;

Claim 28, line 1: "17" should read –27–;

Claim 36, line 3: "15" should read –18–.

Signed and Sealed this

Eighth Day of February, 2000

Attest:

Marcia L. Campbell-Jones

Attesting Officer

Q. TODD DICKINSON

Commissioner of Patents and Trademarks